PARASITES AND SKELETAL INDICATORS OF ANEMIA IN THE EASTERN UNITED STATES

By

Erin Dinneen

RECOMMENDED:

Brian Hemphill, PhD

Carrin Halffman, PhD

Jamie Clark, PhD
Advisory Committee Chair

Ben Potter, PhD
Chair, Department of Anthropology

APPROVED:

Todd Sherman, MFA
Dean, College of Liberal Arts

John Eichelberger, PhD
Dean of the Graduate School

12/7/14
Date

PARASITES AND SKELETAL INDICATORS OF ANEMIA IN THE EASTERN UNITED STATES

A

THESIS

Presented to the Faculty

of the University of Alaska Fairbanks

in Partial Fulfillment of the Requirements

for the Degree of

MASTER OF ARTS

By

Erin E. Dinneen, B.S.

Fairbanks, Alaska

December 2014

Abstract

The goal of this research is to examine the influence of parasitic infection and diet in the etiology of anemia in prehistoric human populations of the eastern United States. Prehistorically, anemia is often attributed to a nutrient-deficient diet, while parasite infection is discussed as a secondary cause if at all. However, parasite infection is a leading cause of anemia in the developing world today. Modern epidemiological studies have demonstrated that parasites thrive or perish under particular environmental conditions, and risk for parasite infection can be predicted based on environment using GIS. Here I apply this method to see whether environmental conditions, acting as a proxy for parasite infection risk, can predict prehistoric skeletal lesion rates for porotic hyperostosis and cribra orbitalia, lesions thought to reflect acquired anemia.

Rates of porotic hyperostosis and cribra orbitalia in the skeletal remains of children and adults were collected from published data for 22 sites in the eastern United States. GIS was used to gather comparable environmental data. Soil drainage, elevation, precipitation, temperature and the surface area of bodies of water were recorded within a 15 km radius of each site. Carbon isotope data deriving from bone collagen and historic hookworm infection rates were also collected when available. Multiple linear regression was used to test how well environmental variables could predict lesion rates.

Statistically significant correlations were found for both adults and children, but the strength and direction of relationships with environmental variables were inconsistent. It is possible that the correlations were related to parasite infection, but it is also possible that the skeletal ‘lesions’ may result from post-mortem bone degeneration rather than anemia. The correlations for porotic hyperostosis and cribra orbitalia were stronger when examined separately

than when examined together, suggesting that the two conditions may have separate etiologies; however, the sample sizes were too small to provide the statistical power required for drawing strong conclusions. Comparison of children and adults showed stronger correlation for children, though when observing the lesions separately this pattern was not consistent. Collagen carbon values and historic hookworm infection rates correlated with lesion rates in children but not adults, perhaps because of differential healing in adults.

These results demonstrate that environmental conditions and skeletal lesions are correlated, but the underlying mechanism for this remains unclear. Larger sample sizes would allow for more robust statistical analyses of the trends observed here. Nevertheless, these results do confirm that porotic hyperostosis and cribra orbitalia cannot be assumed to be the result of nutrient-deficient diets. Interpretation of skeletal data for assessing health in the past must also consider the natural and social context in which individuals lived.

Table of Contents

Page

Signature Page i

Title Page iii

Abstract v

Table of Contents vii

List of Figures xiii

List of Tables xix

List of Appendices xxi

Acknowledgements xxiii

1. INTRODUCTION 1

2. BACKGROUND 5

   2.1 Porotic hyperostosis and cribra orbitalia 5

   2.2 Anemia 11

   2.3 Underlying causes of vitamin deficiencies 13

   2.4 Parasites in the past 14

   2.5 Parasites and GIS today 15

   2.6 Examining diet in the past 16

   2.7 Summary 18

   2.8 Hypotheses 18

3. MATERIALS 21

   3.1 Study Area 21

   3.2 Sites 23

      3.2.1 Irene Mound 24

Page

3.2.2 Monongahela 26

3.2.3 Pearson 27

3.2.4 Sunwatch 27

3.2.5 Mount Nebo 28

3.2.6 Etowah 28

3.2.7 Toqua 29

3.2.8 Ledford Island 30

3.2.9 Averbuch 30

3.2.10 Cox 31

3.2.11 Hardin Village 32

3.2.12 Tinsley Hill 32

3.2.13 Norris Farms #36 33

3.2.14 Lewis Creek Mound 34

3.2.15 Juhle Ossuaries 35

3.2.16 Boytt's Field 35

3.2.17 Ward Place 36

3.2.18 Buffalo 36

3.2.19 Moundville 37

3.2.20 Eiden 38

3.2.21 Anderson 38

3.2.22 East St. Louis Stone Quarry 39

3.2.23 Kane Mounds 39

3.2.24 Summary 40

Page

3.3 Parasites 41

3.3.1 Necator americanus (hookworm) 41

3.3.2 Ascaris lumbricoides (giant roundworm) 44

3.3.3 Trichuris trichiura (whipworm) 45

3.3.4 Giardia lamblia 46

3.3.5 Diphyllobothrium spp. (fish tapeworm) 48

3.3.6 Echinostoma spp. 49

3.3.7 Summary 50

3.4 GIS data 51

3.5 Historic data 52

4. METHODS 53

4.1 Data gathering and entry procedures 53

4.1.1 Soil 55

4.1.2 Elevation 56

4.1.3 Temperature 56

4.1.4 Precipitation 57

4.1.5 Surface area of major lakes and rivers 58

4.1.6. Summary of variables 58

4.2 Statistical procedures 59

4.2.1 Linear regression 60

4.2.2 Assessment of subgroup homogeneity 61

4.2.3 Creating a regression model 65

4.3 Statistical hypotheses 66

Page
5. RESULTS 73
5.1 Regression for adults 73
5.2 Regression for children 76
5.3 Regressions by lesion type 78
5.3.1 Regression for porotic hyperostosis in children 79
5.3.2 Regression for cribra orbitalia in children 79
5.3.3 Regression for porotic hyperostosis in adults 82
5.3.4 Regression for cribra orbitalia in adults 82
5.4 Correlation with $\delta^{13}C$ 85
5.4.1 Adults and $\delta^{13}C$ 85
5.4.2 Children and $\delta^{13}C$ 85
5.5 Comparison of prehistoric and historic data 87
5.5.1 Adult prehistoric lesion rate and historic infection rate 87
5.5.2 Child prehistoric lesion rate and historic infection rate 88
5.5.3 Historic infection rate and environmental predictors 88
5.6 Summary 91
6. DISCUSSION 93
6.1 General patterns observed 93
6.2 Comparison of results to hypotheses 97
6.3 Summary 100
6.4 Limitations 101
6.5 Future research 105
7. CONCLUSION 107

Page
References..109

Appendices 121

List of Figures

Page

Figure 1. Location of sites used in analysis. 25

Figure 2. Lesion rates for each site used in analysis. 26

Figure 3. Scatterplots of relationships between adult lesion rate and environmental variables. . 74

Figure 4. Plot of predicted adult lesion rate against the observed. 75

Figure 5. Scatterplots of relationships between child lesion rate and environmental variables. . 77

Figure 6. Plot of predicted child lesion rate against the observed. 78

Figure 7. Plot of predicted child porotic hyperostosis rate against the observed. 80

Figure 8. Plot of predicted child cribra orbitalia rate against the observed. 81

Figure 9. Plot of predicted adult porotic hyperostosis rate against the observed. 83

Figure 10. Plot of predicted adult cribra orbitalia rate against the observed. 84

Figure 11. Relationship between adult lesion rate and $\delta^{13}C$ average.. 86

Figure 12. Relationship between child lesion rate and $\delta^{13}C$ average.. 87

Figure 13. Relationship between adult lesion rate and historic infection rate. 89

Figure 14. Relationship between child lesion rate and historic infection rate. 90

Figure 15. Plot of predicted historic infection rate against the observed.................................... 91

Figure 16. Soil drainage at Anderson. 122

Figure 17. Soil drainage at Averbuch. 123

Figure 18. Soil drainage at Boytt's Field.. 124

Figure 19. Soil drainage at Buffalo.. 125

Figure 20. Soil drainage at Cox. 126

Figure 21. Soil drainage at Eiden... 127

Figure 22. Soil drainage at East St. Louis Stone Quarry. 128

Figure 23. Soil drainage at Etowah. 129

Figure 24. Soil drainage at Hardin Village. 130

Figure 25. Soil drainage at Irene Mound. 131

Figure 26. Soil drainage at Kane Mounds. 132

Figure 27. Soil drainage at Ledford Island. 133

Figure 28. Soil drainage at Lewis Creek Mound. 134

Figure 29. Soil drainage at Monongahela. 135

Figure 30. Soil drainage at Moundville. 136

Figure 31. Soil drainage at Juhle Ossuary. 137

Figure 32. Soil drainage at Mount Nebo. 138

Figure 33. Soil drainage at Norris Farms #36. 139

Figure 34. Soil drainage at Pearson. 140

Figure 35. Soil drainage at Sunwatch. 141

Figure 36. Soil drainage at Tinsley Hill. 142

Figure 37. Soil drainage at Toqua. 143

Figure 38. Soil drainage at Ward Place. 144

Figure 39. Temperature at Anderson. 145

Figure 40. Temperature at Averbuch. 146

Figure 41. Temperature at Boytt's Field. 147

Figure 42. Temperature at Buffalo. 148

Figure 43. Temperature at Cox. 149

Figure 44. Temperature at Eiden. 150

Figure 45. Temperature at East St. Louis Stone Quarry. 151

Figure 46. Temperature at Etowah. 152

Figure 47. Temperature at Hardin Village. 153

Figure 48. Temperature at Irene Mound. 154

Figure 49. Temperature at Kane Mounds. 155

Figure 50. Temperature at Ledford Island. 156

Figure 51. Temperature at Lewis Creek Mound. 157

Figure 52. Temperature at Monongahela. 158

Figure 53. Temperature at Moundville. 159

Figure 54. Temperature at Juhle Ossuary. 160

Figure 55. Temperature at Mount Nebo. 161

Figure 56. Temperature at Norris Farms #36. 162

Figure 57. Temperature at Pearson. 163

Figure 58. Temperature at Sunwatch. 164

Figure 59. Temperature at Tinsley Hill. 165

Figure 60. Temperature at Toqua. 166

Figure 61. Temperature at Ward Place. 167

Figure 62. Elevation at Anderson. 168

Figure 63. Elevation at Averbuch. 169

Figure 64. Elevation at Boytt's Field. 170

Figure 65. Elevation at Buffalo. 171

Figure 66. Elevation at Cox. 172

Figure 67. Elevation at Eiden. 173

Figure 68. Elevation at East St. Louis Stone Quarry. 174

Figure 69. Elevation at Etowah. 175

Figure 70. Elevation at Hardin Village. 176

Figure 71. Elevation at Irene Mound. 177

Figure 72. Elevation at Kane Mounds. 178

Figure 73. Elevation at Ledford Island. 179

Figure 74. Elevation at Lewis Creek Mound. 180

Figure 75. Elevation at Monongahela. 181

Figure 76. Elevation at Moundville. 182

Figure 77. Elevation at Juhle Ossuary. 183

Figure 78. Elevation at Mount Nebo. 184

Figure 79. Elevation at Norris Farms #36. 185

Figure 80. Elevation at Pearson. 186

Figure 81. Elevation at Sunwatch. 187

Figure 82. Elevation at Tinsley Hill. 188

Figure 83. Elevation at Toqua. 189

Figure 84. Elevation at Ward Place. 190

Figure 85. Precipitation at Anderson. 191

Figure 86. Precipitation at Averbuch. 192

Figure 87. Precipitation at Boytt's Field. 193

Figure 88. Precipitation at Buffalo. 194

Figure 89. Precipitation at Cox. 195

Figure 90. Precipitation at Eiden. 196

Figure 91. Precipitation at East St. Louis Stone Quarry. 197

Figure 92. Precipitation at Etowah. 198

Figure 93. Precipitation at Hardin Village. 199

Figure 94. Precipitation at Irene Mound. 200

Figure 95. Precipitation at Kane Mounds. 201

Figure 96. Precipitation at Ledford Island. 202

Figure 97. Precipitation at Lewis Creek Mound. 203

Figure 98. Precipitation at Monongahela. 204

Figure 99. Precipitation at Moundville. 205

Figure 100. Precipitation at Juhle Ossuary. 206

Figure 101. Precipitation at Mount Nebo. 207

Figure 102. Precipitation at Norris Farms #36. 208

Figure 103. Precipitation at Pearson. 209

Figure 104. Precipitation at Sunwatch. 210

Figure 105. Precipitation at Tinsley Hill. 211

Figure 106. Precipitation at Toqua. 212

Figure 107. Precipitation at Ward Place. 213

Figure 108. Lakes and rivers at Anderson. 214

Figure 109. Lakes and rivers at Averbuch. 215

Figure 110. Lakes and rivers at Boytt's Field. 216

Figure 111. Lakes and rivers at Buffalo. 217

Figure 112. Lakes and rivers at Cox. 218

Figure 113. Lakes and rivers at Eiden. 219

Figure 114. Lakes and rivers at East St. Louis Stone Quarry. 220

Figure 115. Lakes and rivers at Etowah. 221

Figure 116. Lakes and rivers at Hardin Village. 222

Figure 117. Lakes and rivers at Irene Mound. 223

Figure 118. Lakes and rivers at Kane Mounds. 224

Figure 119. Lakes and rivers at Ledford Island. 225

Figure 120. Lakes and rivers at Lewis Creek Mound. 226

Figure 121. Lakes and rivers at Monongahela. 227

Figure 122. Lakes and rivers at Moundville. 228

Figure 123. Lakes and rivers at Juhle Ossuary. 229

Figure 124. Lakes and rivers at Mount Nebo. 230

Figure 125. Lakes and rivers at Norris Farms #36. 231

Figure 126. Lakes and rivers at Pearson. 232

Figure 127. Lakes and rivers at Sunwatch. 233

Figure 128. Lakes and rivers at Tinsley Hill. 234

Figure 129. Lakes and rivers at Toqua. 235

Figure 130. Lakes and rivers at Ward Place. 236

List of Tables

Page
Table 1. Sites used in analysis. 22

Table 2. Anemia-causing parasites and associated ecological variables. 42

Table 3. Chi-squared results for males and females by lesion presence. 64

Table 4. Chi-squared results for adults and children by lesion presence. 64

Table 5. Ecological variables and their expected relationships with PH and CO. 68

Table 6. Results of multiple linear regression with transformed adult lesion rates. 75

Table 7. Results of multiple linear regression for children, five environmental variables. 77

Table 8. Results of multiple linear regression for porotic hyperostosis in children. 80

Table 9. Results of multiple linear regression for cribra orbitalia in children. 81

Table 10. Results of multiple linear regression for porotic hyperostosis in adults. 83

Table 11. Results of multiple linear regression for cribra orbitalia in adults. 84

Table 12. Results of multiple linear regression for historic hookworm infection. 90

Table 13. Summary of relationship directions in multiple linear regression. 95

Table 14. Summary of general hypotheses, results and conclusions. 101

Table 15. Human data used for statistical analysis. 238

Table 16. Environmental data used for statistical analysis. 240

List of Appendices

Page

APPENDIX A Maps for each environmental variable at each archaeological site....................121

A1. Soil Drainage..122

A2. Temperature 145

A3. Elevation 168

A4. Precipitation 191

A5. Surface Area of Major Lakes and Rivers...214

APPENDIX B Raw data used for analysis 237

APPENDIX C Linear regression output from SPSS 241

C1. Multiple linear regression, adults, five environmental variables 242

C2. Multiple linear regression, children, five environmental variables.......................246

C3. Multiple linear regression, PH in children, four environmental variables 250

C4. Multiple linear regression, CO in children, four environmental variables............253

C5. Multiple linear regression, PH in adults, four environmental variables...............256

C6. Multiple linear regression, CO in adults, four environmental variables 259

C7. Simple linear regression for adults and $\delta^{13}C$...262

C8. Simple linear regression for children and $\delta^{13}C$ 264

C9. Simple linear regression for adults and historic infection rates 265

C10. Simple linear regression for children and historic infection rates.......................266

C11. Multiple linear regression, historic infection rates, four environmental variables.........267

APPENDIX D Spearman’s correlation output from SPSS..271

D1. Spearman’s correlation, adults and $\delta^{13}C$..272

D2. Spearman’s correlation, children and $\delta^{13}C$ 273

Acknowledgements

This thesis would not have been possible without help from a number of people. First and foremost I would like to thank my committee members, Dr. Jamie Clark, Dr. Carrin Halffman and Dr. Brian Hemphill. Without their guidance and knowledge I could not have completed this project, and I am grateful for the time and energy that they put into instructing me. I am also indebted to my fellow graduate students, friends and family for their cheer and support throughout this process.

1. INTRODUCTION

Anemia is represented in the skeletal record by small holes in the skull bones called porotic hyperostosis (PH) and cribra orbitalia (CO). In the past and occasionally still today, nutrient deficiencies associated with maize intensive diets were cited as a primary cause of this anemia, with the effects of parasitic infection presented as a secondary factor if mentioned at all (e.g. El-Najjar et al., 1976; Goodman et al., 1984; Rose et al., 1984, 1991; Papathanasiou, 2005).

However, parasitic infection has been demonstrated as an important cause of anemia in developing countries today (Stoltzfus et al., 1997). Subsequent anthropological studies have explored the role of sedentism (Kent, 1986; Reinhard, 1992) and environmental setting (Blom et al., 2005; Pechenkina and Delgado, 2006) in determining risk for parasite infection. It is with these later studies in mind that this thesis seeks to explore the relationship between porotic hyperostosis and cribra orbitalia and parasite infection in the eastern United States. A nutrient deficient diet in tandem with parasitic infection is even more likely to lead to anemia, and so diet will be considered as well. Ecological variables will be used as a proxy for parasite infection risk, and $\delta^{13}C$ values will be used to quantify maize intake.

Studies like Blom et al. (2005), which compared anemia rates among coastal inhabitants of Peru, have provided valuable insights into the role that environment can play in human health. Among other conclusions, Blom et al. found that individuals with anemia buried at lower altitudes and closer to the coast had lower childhood mortality rates. However, studies like this often rely upon subjective categories like “less arid” and “lower altitudes” (Blom et al., 2005, pp 166–167) that can make results tricky to compare and reproduce. The use of GIS to quantify environmental variables can help mitigate this problem. This was well demonstrated in the southeastern United States by Anderson and Allen (2011). The authors found a correlation between historic hookworm infection rates and soil drainage data that were collected using GIS – specifically, infection was more prevalent in counties with more sandy, well-drained soil.

The findings of Anderson and Allen (2011) are important, but they demonstrate a relationship between a single environmental variable which is linked to a single type of parasitic infection. However, cribra orbitalia and porotic hyperostosis are not the result of infection with a single type of parasite; they are instead the result of anemia that is potentially related to multiple parasites. In fact, co-infection with multiple parasites is common and may have a synergistic effect on health, increasing an individual's chances of developing anemia (Pullan and Brooker, 2008). It is for this reason that this investigation will consider multiple parasites and multiple ecological variables.

As previously mentioned, the general hypothesis being tested here is that parasitic infection was a significant cause of anemia in the prehistoric eastern United States. I will do this using multiple linear regression to see whether a combination of environmental variables can predict rates of cribra orbitalia and porotic hyperostosis at the 22 sites considered in this analysis. Comparisons will be made between adults and children, males and females, and porotic hyperostosis and cribra orbitalia separately. Further, I will examine the influence of diet through stable isotopes, and compare the prehistoric lesion rates to historic hookworm infection rates.

Expectations are as follows: (1) lesion rates and environmental variables will demonstrate statistically significant correlation; (2) lesion rates in children will correlate better than those in adults, both since children are more susceptible to parasite infection and because adults may demonstrate differential healing of lesions formed in childhood; (3) porotic hyperostosis and cribra orbitalia will show stronger correlation with the environmental variables when these lesions are separated since it has been proposed that they sometimes have separate etiologies; (4)

lesions in females will correlate better than lesions in males because pregnancy and breastfeeding make women more susceptible to vitamin deficiencies leading to anemia; (5) lesion rates and $\delta^{13}C$ values will correlate and demonstrate a positive relationship, since higher $\delta^{13}C$ values mean more maize consumption and diet is likely a co-contributor with parasite infection to the development of anemia; and (6) prehistoric lesion rates and historic infection rates will be similar since these prehistoric and historic populations lived in the same ecological settings and were therefore exposed to similar risk for parasite infection.

Following this introduction, Chapter 2 will describe background and previous research on porotic hyperostosis and cribra orbitalia and their etiology. The physiological and underlying causes of anemia are also described, as well as the parasites that can lead to anemia and were present in prehistoric North America. The use of GIS to look at modern parasite prevalence will also be reviewed. Finally, Chapter 2 will also discuss the use of stable isotopes to look at diet in the past. Chapter 3 presents the materials used for this analysis, including archaeological sites, parasites, ecological data and comparative historic data. The methods used to collect this data and the statistical hypotheses being tested are presented in Chapter 4. Chapter 5 describes the results of statistical analysis, and Chapter 6 discusses the implications of the patterns observed, limitations of this study and possibilities for future research. Conclusions are presented in Chapter 7.

2. BACKGROUND

This investigation requires an understanding of topics in the fields of archaeology, histology, epidemiology, and stable isotope analysis. Below I will discuss the appearance and identification of porotic hyperostosis and cribra orbitalia, and how they form as a result of anemia. I will then outline the various causes of anemia, and the underlying reasons for vitamin-deficiency anemia, including dietary deficiency and parasitic infection. Next I will discuss how parasites are identified archaeologically, and how GIS has been used to examine parasite infection both in the past and the present. I will then describe how stable isotopes can be used to learn about diet in the past. Finally, I will outline the general hypotheses for this analysis based on this background information.

2.1 Porotic hyperostosis and cribra orbitalia

Porotic hyperostosis and cribra orbitalia describe the widening of the diploe and thinning of the outer table of the cranium, resulting in tiny foramina in the cortical layer. When these foramina appear on the cranial vault, usually on the frontal, parietal and occipital bones, it is called porotic hyperostosis. When they appear on the orbital roof, it is called cribra orbitalia (Stuart-Macadam, 1992). These lesions are seen on both juvenile and adult crania, though it has been suggested by many that they are the result of childhood episodes of illness (Stuart-Macadam, 1985; Perry, 2005; Walker et al., 2009). This is because these lesions appear to be less ‘active’ in adults than children (i.e. more healing has taken place in adult crania).

Early identifications of porotic hyperostosis and cribra orbitalia relied upon gross observation of the skull. There are a number of published standards for visually collecting information from skeletal remains, and one of the most commonly used is Buikstra and

Ubelaker's (1994) *Standards for Data Collection from Human Skeletal Remains*. Scoring with this method includes the severity of the porosity (very indistinct, true, coalescing, or coalescing with expansive changes) as well as whether the lesions are active or healed (Buikstra and Ubelaker, 1994:121). These stages are described as well as illustrated with pictures. Methods such as this are and will likely continue to remain popular because they are relatively easy to learn, inexpensive and quick (Grauer, 2008). However, visual assessment does have clear limitations when it comes to interobserver error, as demonstrated in a study by Jacobi and Danforth (2002). The authors found that both experienced and inexperienced researchers demonstrated relatively high (>80%) levels of agreement when lesions were present, though the scoring of the severity of these lesions was less consistent. However, when lesions were in fact absent, scorers often mismarked them as present, suggesting that expectations and preconceptions had an influence on observations.

One of the most commonly cited causes of porotic hyperostosis and cribra orbitalia is anemia (e.g. Lovell, 1997; Salvadei et al., 2001; Blom et al., 2005). In an early investigation focused on porotic hyperostosis, Angel (1966) examined a large sample of skeletons from the Mediterranean including Greece, Turkey and Cyprus. The author observed that groups living close to marshes had higher rates of porotic hyperostosis than those living in drier areas. He concluded that the lesions were the result of inherited anemias (thalassemia or sickle cell) which rose in frequency as increased resistance to malaria became more evolutionarily advantageous.

However, in most populations, inherited anemias are not as common as the frequency of porotic hyperostosis and cribra orbitalia would seem to indicate. An alternative explanation is chronic iron deficiency anemia, which is today one of the most commonly cited cause of porotic hyperostosis and cribra orbitalia. Early proponents of this were Carlson and coworkers (1974) who proposed iron deficiency anemia as the cause of porotic cranial lesions in Meriotic, X-group and Christian skeletons from ancient Nubia. The authors observed higher occurrences of porotic hyperostosis and cribra orbitalia in the youngest and oldest portions of the population, and they turned to archaeological and ethnographic evidence to explain this. They observed that diet during all time periods would have been primarily composed of milled cereals like millet and wheat which contain little iron. A similar diet has been reported in this area in modern times, and common too is iron deficiency anemia. Parasitic and bacterial infections are also common, leading to blood loss and malnutrition. This would likely have been the case in the past as well, and older individuals and children would probably have been most susceptible to illness and malnutrition related to poor diet and parasitic infection. Based on these factors, the authors suggest that in prehistoric Nubia, cribra orbitalia and porotic hyperostosis were the result of acquired iron deficiency anemia rather than hereditary anemia. Further, the authors observe that this diagnosis fits in well with clinical research demonstrating that iron deficiency anemia can result in porotic cranial lesions in modern populations.

Hereditary disease is also unlikely to be at the root of anemia in the pre-Columbian New World – the crossing of the Bering land bridge would have meant prolonged exposure to cold, which would have killed the mosquito vectors of malaria. Though there is evidence for the pre-Columbian presence of mosquito vectors in Peru, it is likely that this was the result of a transpacific migration, and it does not appear that malaria was common in the pre-Columbian New World (Drake and Oxenham, 2013). This means that there was no incentive for selection in favor of genetic traits like sickle cell and thalassemia which make it difficult for some forms of malaria to enter the body.

Early advocates for iron deficiency anemia as a cause of porotic hyperostosis and cribra orbitalia in the New World were El-Najjar et al. (1976) who looked at these lesions in two ecological zones in the American Southwest, comparing people who lived in canyon bottoms to those who lived on the sage plain. The authors examined five sites from these ecozones and observed that while 54.1% of canyon inhabitants had cribra orbitalia or porotic hyperostosis, only 14.5% of sage plain inhabitants did. Archaeological and historical evidence suggests that the canyon dwellers were highly dependent on maize, which grew well there, and storage of this grain allowed for population expansion in an otherwise somewhat marginal environment. In contrast, sage plain dwellers had significantly more access to wild animals, though they also supplemented their diet with agriculture. Based on these observations, the authors suggested that the canyon dwellers' dependence on iron-poor maize resulted in higher rates of iron deficiency than the more meat-focused diet at the sage plain sites. They proposed that this iron deficiency was a primary cause of anemia leading to porotic hyperostosis in the past.

However, as in prehistoric Nubia, diet is not the only proposed cause of anemia in the American Southwest. Kent (1986) studied the prehistoric Anasazi of this region, focusing in particular on the cultural and economic context in which these people lived. Using ethnographic and historic evidence, Kent suggested that the Anasazi were not in fact as maize dependent as many suggested, and that events like the exchange of maize for meat with more mobile groups could have depressed evidence for meat consumption in archaeological contexts. Further, the evidence for prehistoric human parasite infection in this area is strong, coming from finds like trematode eggs in coprolites from the site of Glen Canyon in Utah. Parasitic infections like these can lead to blood loss and diarrhea, as well as increased susceptibility to other infections. With the increased sedentation and population aggregation brought about by intensive maize agriculture, the chances of encountering and contracting infection would have increased significantly. This is demonstrated in modern ethnographic and clinical literature, and when Kent (1986) compared rates of porotic hyperostosis and cribra orbitalia in children in the prehistoric Anasazi to that of modern developing countries, the rates of infection appeared very similar. From this the author concluded that Anasazi diet was probably nutritionally adequate, and was "virtually inconsequential in the rise in frequency of porotic hyperostosis and chronic iron deficiency anemia" (Kent, 1986, p 605). Iron deficiency anemia was instead proposed to be a result of increased parasitism and infection due to sedentism and increased population size.

Porotic hyperostosis and cribra orbitalia have also been associated with rickets, scurvy, or other inflammatory processes on the cranium or scalp (Schultz et al., 2001). These can sometimes be macroscopically differentiated from lesions due to anemia based on the involvement of particular other skeletal elements; they can often be microscopically differentiated because the body reacts variably to diseases, leading to variation in microscopic bony changes. Schultz (2001) used thin-sectioning in combination with light microscopy to look at the microscopic changes in bone structure that occur with various diseases. Using one modern and four archaeological individuals from Germany, Turkey and Florida who had been microscopically diagnosed with anemia, Schultz (2001) illustrates thin sections that represent the bony changes associated with anemia. Specifically, these morphological features are thinning of the external lamina and enlargement of the cancellous bone; these usually occur together. These illustrations were then compared to known cases of other diseases to highlight the differences. Microscopic analysis of one individual from the Sundown Site in Arizona demonstrated that what macroscopically appeared to be porotic hyperostosis was in fact the result of a subperiosteal hematoma due to chronic scurvy. Analysis of an individual from the İkiztepe site in Turkey demonstrated that porotic lesions that macroscopically suggested anemia were in fact the result of rickets.

Post-depositional destructive processes can also result in porosities that resemble porotic hyperostosis and cribra orbitalia. Wapler et al. (2004) also used thin-sectioning to look at the etiology of cribra orbitalia in 85 prehistoric individuals from northern Sudan. Analysis of the bone using polarized light, which allows one to see bone collagen and therefore lamellar orientation and histologic structure, showed that anemia was responsible in only 43.5 % of cases. Other conditions like osteitis and hypervascularization accounted for cribra orbitalia in 36.5% of individuals, and fully 20% of the sample showed cribra orbitalia as a result of postmortem erosion. Changes due to postmortem erosion were identified through the disintegration of collagen fibers in combination with no features of bone reaction.

Non-destructive methods have also been applied to the study of porotic hyperostosis and cribra orbitalia, including radiographs, histological studies, and computed tomography (Stuart-Macadam, 1987; Vasalech, 2011; Galea, 2013). These studies all demonstrated that while the macroscopic, external appearance of porotic hyperostosis and cribra orbitalia can be indicative of anemia, this is not always the case. Like thin-sectioning, these studies showed that lesions could also result from trauma, inflammation and subperiosteal hemorrhage.

Studies like these have demonstrated that porotic hyperostosis and cribra orbitalia are not exclusively the result of anemia. It is unfortunate that these kinds of approaches are often seen as either too time-consuming, destructive or expensive to be widely used (Grauer, 2008). Despite other possible etiologies, cribra orbitalia and especially porotic hyperostosis are still in many studies assumed to be the result of some form of anemia. This is probably due to a combination of the expense and time required for microscopic analysis and the long-standing tradition in the field of interpreting these lesions as an expression of anemia. Though this analysis will follow this convention and depend upon this assumption, any results should be interpreted with caution given the variety of possible etiologies for these lesions.

Since they are often reported together in the literature, I will consider both porotic hyperostosis and cribra orbitalia together in my overall sample. However, these lesions have a relationship that is still a matter of debate. As indicated by Angel's (1966) terminology, they have long been thought to share a common etiology, but recently researchers such as Walker and coworkers (2009) and Rothschild (2012) have proposed that they may not. This is because while porotic hyperostosis appears to have a somewhat limited number of causes, cribra orbitalia can result from any subperiosteal inflammation, even including inflammation resulting from traumatic injury. It is for this reason that I will examine correlations with environmental variables for these two lesions separately as well as in combination, though this was not possible for each site since some investigators did not report them separately.

2.2 Anemia

As previously mentioned, bioarchaeologists have long argued that porotic hyperostosis and cribra orbitalia were primarily caused by iron deficiency anemia. However, more recent work has shown that anemia can result from a number of other processes (Walker et al., 2009; Gowland and Western, 2012; Rothschild, 2012). Convincing cases have been made for anemias associated with other conditions, particularly deficiencies such as folic acid or vitamins B_{12}, E and A (Oski and Barness, 1967; Semba and Bloem, 2002; Stabler and Allen, 2004). When combined with diseases like malaria, these deficiencies are particularly likely to lead to severe anemia (Gowland and Western, 2012).

Anemia is a condition under which the body has an insufficient supply of hemoglobin, usually associated with a reduction in circulating red blood cells (RBCs). RBCs and hemoglobin are essential for transporting oxygen throughout the body. Anemia can result from inherited hemoglobin or red cell defects like sickle cell or thalassemia, which usually cause reduced survival of RBCs. It can also be acquired, through a decrease in RBC production (often the result of vitamin deficiencies or marrow replacement), an increase in RBC destruction (often the result of infections or vitamin deficiencies), or as the result of chronic disease (Seeber and Shander, 2012; Mehta and Hoffbrand, 2013).

The body responds to reduced oxygen flow in a number of ways, and short-term reductions in RBCs or hemoglobin can often be compensated through mechanisms like increased oxygen extraction by tissues and redistribution of blood flow to organs with high oxygen demand (Seeber and Shander, 2012). In long-term anemia, however, the body responds by causing marrow to expand (hypertrophy) to create more red blood cells. This happens especially in the primary blood production centers in the body, which for children are in the cranial vault and the medullary cavities of the long bones. When marrow hypertrophy is severe enough it reabsorbs some of the ectocranial surface, resulting in tiny foramina in the bone (Walker et al., 2009).
The reason that iron deficiency anemia has recently been doubted as a cause for porotic hyperostosis and cribra orbitalia has to do with the way human bodies respond to this particular deficiency. Walker et al. (2009) propose that while iron deficiency anemia causes inefficient RBC production, it does not involve massive RBC destruction. The inefficient RBC production caused by iron deficiency could actually prevent RBC production at a volume large enough to cause severe marrow hypertrophy. Vitamin B_{12} deficiency, however, does cause severe marrow expansion (Walker et al., 2009; Rothschild, 2012). However, Oxenham and Cavill (2010) point

out that not all clinical literature supports this view, and suggest that dismissal of iron deficiency as a potential cause of anemia would be hasty. It is worth noting that vitamin B_{12} deficiency frequently co-occurs with iron deficiency, since the process of marrow hypertrophy consumes vitamins like iron. In these cases iron deficiency is a result of marrow hypertrophy, not the cause (Rothschild, 2012). This reinforces the fact that iron and vitamin B_{12} deficiency are closely related phenomena. Teasing these apart is beyond the scope of this project; the aim is rather to examine the roles of parasitic infection and diet in causing nutrient deficiencies that lead to anemia.

2.3 Underlying causes of vitamin deficiencies

There are two underlying causes of iron and vitamin B_{12} deficiency – inadequate intake and improper absorption. While iron and B_{12} intake can decrease periodically for most adults and not be a problem (stores in the body are depleted fairly slowly), low intake is a greater concern for mothers who are nursing or pregnant. Pregnancy and nursing deplete iron and vitamin B_{12} stores more quickly, and inadequate intake can lead to a baby being born with severe vitamin deficiencies (Stabler and Allen, 2004; Walker et al., 2009). The primary sources of vitamin B_{12} are animal products, including meat, eggs, and dairy (Olivares et al., 2002; Stabler and Allen, 2004). Iron can come from a wider range of food sources, but the presence of iron absorption inhibitors like phytate and polyphenols in many plants means that the bioavailability of iron from these sources is actually very low (Tatala et al., 1998). Animal products therefore remain the primary sources of both iron and vitamin B_{12} for humans.

However, inadequate absorption can cause iron and vitamin B_{12} deficiency even if sufficient amounts are consumed (Tatala et al., 1998; Stabler and Allen, 2004; Stoltzfus et al., 2004). A common cause of malabsorption is parasitic infection, including *Necator americanus, Trichuris trichiura*, *Ascaris lumbricoides*, *Diphyllobothrium latum*, and *Giardia lamblia* (Farid et al., 1969; Cox, 1993; Olivares et al., 2002; Carvalho-Costa et al., 2007; Scholz et al., 2009). Some parasites actually absorb the iron and vitamin B_{12} that the host consumes, while others cause diarrhea which inhibits the absorption of nutrients in general. Blood loss is also often associated with parasite infection, and when bled chronically, laboratory rats demonstrate significantly more marrow expansion than those fed an iron-deficient diet (Burkhard et al., 2001). Parasitic infections are especially likely to lead to anemia when they are combined with low intake of iron and vitamin B_{12} (Stoltzfus et al., 2004; Walker et al., 2009). Parasitic infection is therefore highly likely to cause or exacerbate the kind of long-term nutrient deficiencies that could cause bone to remodel.

2.4 Parasites in the past

Parasites are recognized archaeologically primarily from the study of desiccated feces, called coprolites. Mummies and latrine soils can also provide insight. The main focus of archaeoparasitology is on arthropods (usually lice) and helminths. Eggs and larvae are most commonly preserved, though particular helminths have a tough outer cuticle which allows for preservation of some adults (Reinhard, 1990). The pre-Columbian presence of various parasites in the New World is still debated, though new finds and advances in genetic techniques are currently expanding our knowledge (Gonçalves et al., 2003; Morgan et al., 2005; Sianto et al., 2005; Reinhard et al., 2013).

While coprolites, latrine soils and mummies can provide excellent information about parasites in the past, each of these requires very particular environmental conditions for preservation. The old saw 'the absence of evidence is not evidence of absence' is as true when it comes to prehistoric parasitism as it is in other areas of archaeology. It is for this reason that environmental variables are potentially valuable as proxies for the likelihood of parasite infection, as in Gowland and Western's (2012) study of malaria in Anglo-Saxon England. The authors collected data on the archaeological occurrence of cribra orbitalia in eastern England and compared this spatial data with the distribution of environmental variables associated with mosquito habitat. Presence of mosquito habitat was inferred through comparison of environment and historically reported cases of malaria and ague. A strong correlation was observed between cribra orbitalia and mosquito habitat, suggesting that cribra orbitalia may have been the result of anemia associated with malaria during this time period.

Correlation between environment and parasitic infection in the past was also demonstrated in a study by Anderson and Allen (2011), who examined historic hookworm infection rates from the southeastern United States and found that infection was more common in counties with sandy, well-drained soils. This study is particularly important because it overlaps the geographic region of interest for this thesis, and it demonstrates that at least historically there was correlation between environmental setting and parasite infection. The present study expands upon this work by considering multiple environmental variables and comparing them to prehistoric rather than historic health data.

2.5 Parasites and GIS today

GIS has been used to predict risk of parasite infection in a number of modern epidemiological studies (Brooker and Michael, 2000; Raso et al., 2005; Bethony et al., 2006). Factors that affect risk of infection are both small and large in scale, and include things like how much contact people have with each other, sanitation practices, treatment availability and use, and environmental variables that affect the life cycle of parasites (Brooker and Michael, 2000). It is this last category that is most useful for broad-scale analyses, and I believe also has the most potential for examining risk of infection in the past. Correlations have been shown in modern studies between environmental variables and frequency of parasite infection because environment plays a key role in parasitic life-cycles. For example, the trematode *Schistosoma mansoni* spends part of its life-cycle using a snail as an intermediate host before infecting its primary host, humans. These snails have fairly well defined minimum and maximum temperatures at which they can survive (between 16°C and 30°C), thus limiting the possible environments in which *S. mansoni* can infect humans. Other factors include distance to water, water velocity and altitude (Brooker and Michael, 2000). It is for this reason that I have chosen to use environmental variables as a proxy for risk for parasite infection. The specific variables used in this study will be discussed in more detail in Chapter 3.

2.6 Examining diet in the past

Many of the vitamins and minerals that humans need to maintain healthy bodies are obtained through diet. As discussed, these include iron and vitamin B_{12}, a lack of which can lead to anemia. Stable isotope analysis can be useful for looking at animal product consumption and C_3 vs. C_4 plants in the diet, providing insight into whether vitamin and mineral deficiencies due to under-consumption were likely.

Since bone remodels throughout life, isotope values from this tissue represent average diet over a person’s lifetime (Hedges et al., 2007). The most frequently studied isotopes are those of carbon and nitrogen (Lee-Thorp, 2008). The ratios of their heavy to light isotopes (expressed

$\delta^{13}C$ and $\delta^{15}N$) can be used to estimate the proportional contribution of various food sources to the diet, including animal versus plant products, and C_3 plants (e.g., wheat) versus C_4 plants (e.g., maize) (Ben-David and Flaherty, 2012).

Collagen is the most commonly analyzed portion of bone and tooth dentine. In North America, one of the earliest anthropological uses of carbon isotope ratios was van der Merwe and Vogel's (1978) study looking at the introduction of maize agriculture in eastern North America. A dramatic shift was seen later isotopically than archaeologically, around AD 1000. Debate over the implications of these findings eventually led to the discovery that dietary protein is preferentially routed to collagen, suppressing indications of starchy foods like maize (Ambrose and Norr, 1993; Tieszen and Fagre, 1993). That is, until maize is a large enough part of someone's diet, it will not appear at all in collagen because its isotopic signature will be 'crowded out' by protein-rich foods.

Since carbohydrates and fats do not contain nitrogen, we can assume that nitrogen isotope ratios of consumer tissues reflects dietary protein sources (DeNiro and Epstein, 1981; Ambrose et al., 1997). Because consumers have higher $\delta^{15}N$ values than the diet they consume, there is an increase in nitrogen isotope ratios as one moves up the food chain (O'Connell et al., 2012). Thus, nitrogen isotope ratios are particularly useful for looking at the consumption of animal products (the primary sources of iron and vitamin B_{12}).

Unfortunately, isotope research in the eastern United States often focuses on agriculture and maize consumption, and therefore $\delta^{13}C$ data from collagen is more widely reported than $\delta^{15}N$. It is for this reason that carbon rather than nitrogen data are used in this analysis. It is possible though that $\delta^{13}C$ will demonstrate positive correlation with lesion rates since higher (less negative) $\delta^{13}C$ values reflect increased maize consumption, and as discussed, maize dependence has been suggested as an important cause of anemia in the past.

2.7 Summary

Porotic hyperostosis and cribra orbitalia are skeletal lesions which are usually attributed to anemia. This anemia must be chronic in order to cause bone to remodel. In North America, this anemia is usually attributed to dietary deficiency in one or more essential nutrients or malabsorption of these nutrients due to parasite infection. Nutrient-deficient diets have been proposed as a result of maize-dependence at a number of sites, and this hypothesis can at least theoretically be tested through stable isotope analysis wherever there are skeletal remains. Parasite infection can be demonstrated through coprolites and mummies, but this is only possible where preservation conditions are appropriate. GIS provides a method for establishing environmental proxies that represent risk of parasite infection, making it possible to consider the likelihood of parasite infection in the past even in places where physical preservation of parasites is not possible. GIS data has the further advantage of being quantitative and reproducible.

2.8 Hypotheses

Based on the background information presented here, I have formulated six general hypotheses. I will describe each and outline the reasoning behind them below.

1) *There will be significant correlation between prehistoric lesion rates and environmental variables*

This would support the idea that parasite infection was an important cause of anemia in the prehistoric eastern United States, and this is likely given the association between parasite infection and anemia in clinical studies in the modern developing world.

2) *Children will show stronger correlation than adults*

This is likely for two reasons. The first is that healing in adults may mean that their lesion rates do not reflect the amount of anemia they experienced as children. The second is that children are more likely to develop chronic anemia from a parasite infection since their bodies do not have the same built-up reserves of iron and B_{12} that adults have.

3) *Women will show stronger correlation than men*

I believe this will be true because pregnant or breastfeeding women have to share the vitamins and minerals that they consume with another body. This means that parasite infection or even a temporary dietary deficiency are more likely to lead to anemia since the body's reserves are depleted more quickly.

4) *Cribra orbitalia and porotic hyperostosis will correlate better separately than when grouped together*

This is likely since separate, non-anemia etiologies have also been proposed for both of these lesions, particularly cribra orbitalia.

5) *$\delta^{13}C$ values will be able to successfully predict prehistoric lesion rates*

It is not proposed here that parasite infection was the only cause of anemia in the prehistoric eastern United States, but that it was an additional important factor alongside diet. Chronic anemia is most likely to result from a combination of malnourishment and parasite infection.

6) *Historic rates of infection and prehistoric lesion rates will be similar*

This is likely since the prehistoric and historic groups of people lived in the same or similar ecological settings, and were therefore exposed to the same risk for parasite infection.

3. MATERIALS

To investigate the hypotheses just outlined, this investigation utilizes both archaeological and environmental data. The latter are based on the requirements of parasites that (1) cause anemia, and (2) were present in the prehistoric New World. I will begin by discussing why I chose the eastern United States as a geographic region, and then describe the sites from which I drew data on prehistoric skeletal lesion rates and stable isotopes. This background information is taken from previous investigations, and so there are some inconsistencies in the data available for each (e.g. paleodemographic profiles have not been developed for every site). Next I will discuss each of the parasites that are considered here in terms of their life cycle, ability to cause vitamin-deficiency anemia, and their presence in the prehistoric New World. Based on this information I will briefly outline the environmental variables used to represent risk for parasite infection, and then describe the historic data that was available to compare with nine of the archaeological sites.

3.1 Study Area

This analysis focuses on the eastern United States for a number of reasons. First, there is a relatively long history of bioarchaeological research in this region, making it a rich source of published data (see Table 1). There is also a solid history of stable isotope analysis in this geographic area (see Table 1). Further, while groups were certainly culturally heterogeneous, agricultural and settlement practices were similar enough that a focus on environmental variables is possible – that is, cultural differences are not so dramatic that they are likely to obscure other potential influences on anemia like dietary or environmental factors. The most obvious way to mitigate cultural differences between groups would be to compare individuals within a single

Table 1. Sites used in analysis.

Site Name	**Location**	***N****	**Source (skeletal lesion data)**	**Stable isotope data**	**Source (stable isotope data)**
Irene Mound	Chatham County, GA	187	Steckel et al., 2002	yes	Larsen et al., 2001
Monongahela	Westmoreland County, PA	121	Steckel et al., 2002	yes	Farrow, 1986
Buffalo	Kanawha County, WV	99	Steckel et al., 2002		
Pearson	Seneca County, OH	95	Steckel et al., 2002	yes	Stothers and Abel, 1989
Sunwatch	Clermont County, OH	128	Steckel et al., 2002	yes	Cook and Schurr, 2009
Boytt's Field	Union County, AR	25	Rose et al., 1991		
Ward Place	Morehouse Parish, LA	25	Rose et al., 1991		
Mount Nebo	Madison Parish, LA	77	Rose et al., 1991		
Anderson	Warren County, OH	44	Lallo, 1979	yes	Cook and Schurr, 2009
Ledford Island	Bradley County, TN	343	Helms, 2012		
Moundville	Perry County, AL	162	Powell, 1991	yes	Schoeninger and Schurr, 1998
East St. Louis Stone Quarry	St. Clair County, IL	41	Milner, 1991	yes	Hedman et al., 2002
Kane Mounds	Madison County, IL	98	Milner, 1991	yes	Buikstra and Milner, 1991
Averbuch	Davidson County, TN	732	Eisenberg, 1991	yes	Schurr, 1992
Norris Farms #36	Fulton County, IL	170	Milner et al., 1991	yes	Buikstra and Milner, 1991
Hardin Village	Greenup County, KY	292	Cassidy, 1972	yes	Broida, 1984
Eiden	Lorain County, OH	122	Lallo and Blank, 1977		
Tinsley Hill	Lyon County, KY	81	Lane, 1993	yes	Schurr and Powell, 2005
Lewis Creek Mound	Augusta County, VA	26	Gold, 2004	yes	Trimble, 1996
Cox	Anderson County, TN	190	Vogel, 2007		
Etowah	Bartow County, GA	125	Blakely, 1980		
Toqua	Monroe County, TN	245	Parham and Scott, 1980		
Juhle Ossuaries	Charles County, MD	208	Chase, 1988		

**N* = number of individuals examined for skeletal lesions group, but as Roux (2004:104) points out:

> Studies that focus on what distinguishes sick individuals from healthy individuals *within* a population or group may miss important disease determinants. This is because population-level factors are invariant within a population and, hence, cannot be investigated in studies restricted to comparisons of individuals within a population.

The appropriate scale for group-level analysis is one that balances the need for the region to be large enough to provide good rate estimates and demonstrate environmental differences, yet small enough to be relatively homogenous in terms of other factors like subsistence practices (Rezaeian et al., 2007). It is for these reasons that I have restricted my investigation in the eastern United States.

3.2 Sites

The sample of sites was limited to those occupied by sedentary groups that practiced agriculture. The occupation dates for these sites range from AD 750 (Mt. Nebo) to AD 1650 (Boytt's Field and Ward Place). The sites from which I drew data and the sources of this information are listed in Table 1. I began my search for published lesion rates with the Global History of Health database (Steckel et al., 2002), a project that was started at Ohio State University. This database contains standardized skeletal data regarding health, age, sex, and context for a total of 12,520 individuals from 65 locations in the Western Hemisphere (Steckel et al., 2002). The project is currently being expanded to include a European module. After collecting data from as many Global History of Health sites as met my criteria, I expanded my search by following references listed in articles discussing potential causes of porotic hyperostosis and cribra orbitalia. More details on the criteria used for selecting sites will be discussed in Chapter 4. Figure 1 is a map showing the location of each site, and Figure 2 shows the rates of porotic hyperostosis and cribra orbitalia for the whole group sample at each site. A brief introduction to each site is presented below. Any inconsistencies in the background information presented are the result of differences in the information presented in the source reports and analyses.

3.2.1 Irene Mound

Irene Mound was located in Chatham County, Georgia, on a bluff on the south bank of the Savannah River. The coast in this area transitions from the mainland to marshy islands, then tidal creeks and sounds before reaching the open Atlantic. The population buried here practiced agriculture and maize was a significant part of their diet, though locally foraged wild foods were also important (Larsen et al., 2002). There are two mounds from which samples derive, the Irene Burial Mound and Irene Large Mound, though there were also a number of smaller mounds on site. The burials date from AD 1150 to 1550 (Williams, 2005). Individuals buried here appear to have closer biological links to inland populations than to the coastally-focused Guale people (Larsen, 2001). The site appears to have been a ceremonial or political center, though there is also some evidence of regular habitation. There is no evidence for contact with Europeans, though Irene Mound seems to have been in use nearly up until the arrival of the Spanish (Caldwell et al., 1941). The skeletal sample used in this analysis consisted of 187 individuals with an overall lesion rate of 2.67% (Steckel et al., 2002). The $\delta^{13}C$ isotopic sample consisted of 10 individuals with an average value of -13.31 (Larsen et al., 2001).

Figure 1. Location of sites used in analysis.

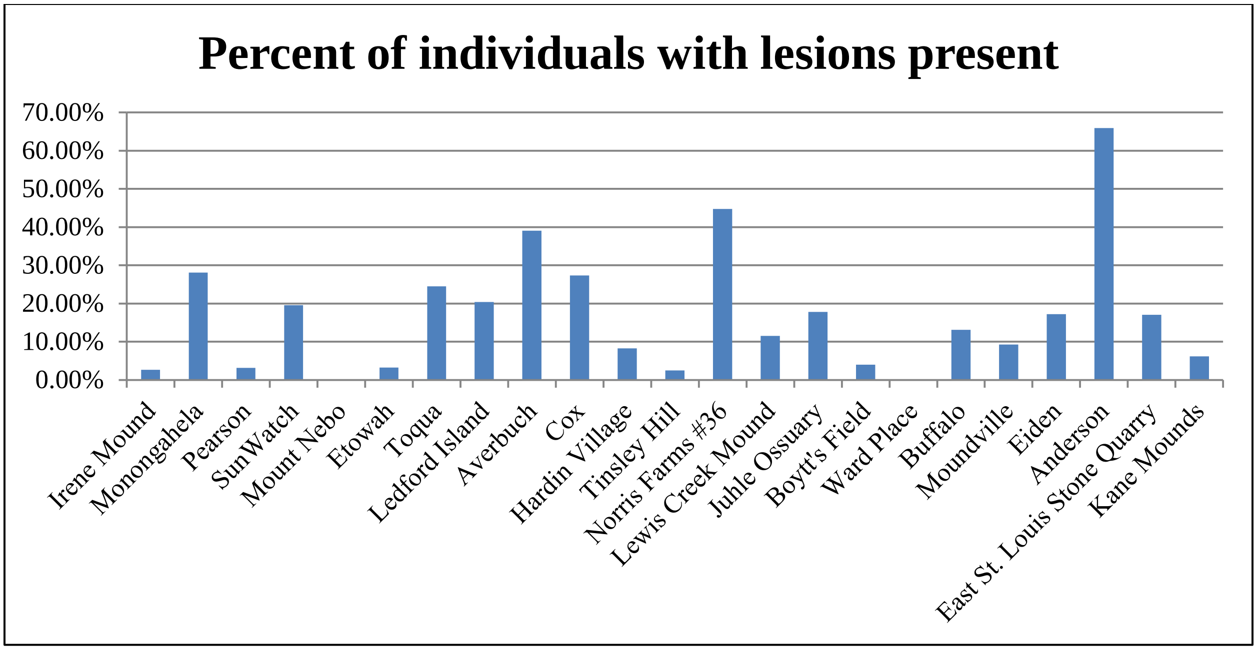

Figure 2. Lesion rates for each site used in analysis.

3.2.2 Monongahela

The Monongahela samples consist of an aggregation of individuals from 50 sites between the Monongahela and Youghiogheny Rivers in southwestern Pennsylvania (Williams, 2005). Maize agriculture was the subsistence focus, though wild resources like nuts and fauna were also important. Villages, hamlets, farmsteads and camps are all represented in this area. The skeletal samples date from AD 1000 to 1630. The age-at-death distribution for this group shows that people aged 0 to 5 and 45+ are under-represented, and Sciulli (2002) suggests that this is the result of some seasonal mobility, poor preservation of the young, and biased adult aging techniques. The skeletal sample probably does not represent everyone who died throughout the year, but instead primarily people who died between late spring and early autumn. The skeletal sample used in this analysis consisted of 121 individuals with an overall lesion rate of 28.1% (Steckel et al., 2002). The δ^{13}C isotopic sample consisted of 11 individuals with an average value of -10.50 (Farrow, 1986).

3.2.3 Pearson

The Pearson Complex consists of three overlapping habitation sites in northern Ohio, located near the Sandusky River. The site is located on a bluff along Green Creek, slightly elevated from the surrounding rolling lake plain. The skeletal samples come from two associated cemeteries from the Eiden (AD 1000-1200) and Fort Meigs (AD 1525-1550) phases of occupation (Stothers and Abel, 1989). Approximately 90% of the skeletons are from the earlier occupation and 10% are from the later. The inhabitants of this site were intensive maize agriculturalists, though hunting and fishing also contributed to their diet. Paleodemographic analysis shows that both younger and older individuals are under-represented in the earlier phase cemetery, and archaeological evidence suggests that occupation at least during this phase may have been restricted to the late spring and summer (Sciulli et al., 1996). The skeletal sample used in this analysis consisted of 95 individuals with an overall lesion rate of 3.16% (Steckel et al., 2002). The $\delta^{13}C$ isotopic sample consisted of 4 individuals with an average value of -12.81 (Stothers and Abel, 1989).

3.2.4 Sunwatch

The Sunwatch site is located in southwestern Ohio and was previously called the Incinerator site. The layout is circular and the site was organized into specialized areas for burials, pit features, houses and a stockade. Burial groups are associated with corporate houses and date from approximately 1150 to 1450 (Cook and Schurr, 2009). Maize agriculture was important, though seasonal mobility may have occurred (Williams, 2005). Maize became particularly important in the latest phases of occupation, and the increase in consumption was accompanied by the emergence of differential burial treatments possibly linked to status differences (Cook and Schurr, 2009). This is in contrast to the majority of the sample from earlier occupation phases, which show little if any status differentiation (Griffin, 1992). The skeletal sample used in this analysis consisted of 128 individuals with an overall lesion rate of 19.53% (Steckel et al., 2002). The $\delta^{13}C$ isotopic sample consisted of 58 individuals with an average value of -11.10 (Cook and Schurr, 2009).

3.2.5 Mount Nebo

The Mount Nebo site is located in northeast Louisiana, on a natural embankment of the Tensas River. The site consists of a single mound built in seven stages, two of which (A and F) contained the skeletons used in this analysis (Kassabaum, 2011). Approximately half of the total sample from Mount Nebo comes from each of these two building phases, and both date to the Coles Creek Period (AD 750-1200) (Rose et al., 1991). Children appear to be underrepresented in this population, either as a result of differential preservation or intentional choice by the original buriers (Kassabaum, 2011). Coles Creek subsistence was based on cultivation of indigenous starchy seeds like knotweed and goosefoot, as well as hunting and gathering (Rose et al., 1991). The skeletal sample used in this analysis consisted of 77 individuals with an overall lesion rate of 0.00% (Rose et al., 1991).

3.2.6 Etowah

The Etowah site is located on a floodplain just north of the Etowah River in northwestern Georgia. It is a Mississippian site occupied from approximately AD 1000 until the early contact period. The inhabitants were maize agriculturalists, and were also well positioned to exploit the wild resources of the piedmont and Appalachian plateau (Blakely, 1980). The site contains both mound (Mound C) and village components, and the former is known for the fine grave goods associated with the burials here. Mound C burials date from approximately AD 1000 to 1400. The village interments date primarily from AD 1400 to 1600 lack elaborate grave goods (Blakely, 1995). There appears to have been social stratification, though whether it was achieved or ascribed is not clear. The samples from both the village and the mound do not include any individuals 15 years of age or younger. There was no statistically significant difference in the age distribution, sex distribution, or frequency of porotic hyperostosis between the village and mound samples (Blakely, 1980); I have therefore combined them into a single sample for this analysis. The skeletal sample used in this analysis consisted of 125 individuals with an overall lesion rate of 3.20% (Blakely, 1980).

3.2.7 Toqua

The Toqua site is located on the southern bank of the Little Tennessee River. It was excavated in the mid-1970s in preparation for the flooding of the new Tellico Reservoir. The site contained two mounds, a village with numerous structures and features, a plaza and multiple palisades (Boyd and Boyd, 1991). The mounds were both built in multiple stages over a long period of time (Schroedl, 1998). The primary occupation lasted from approximately AD 1300 to 1550 and the inhabitants of Toqua were sedentary, intensive maize agriculturalists (Parham and Scott, 1980). Social stratification was present and mound burials appear to be higher-status individuals; however, there was not a statistically significant difference in frequency of porotic hyperostosis between mound and village (Parham and Scott, 1980), and I therefore consider these groups together in this analysis. The skeletal sample used in this analysis consisted of 245 individuals with an overall lesion rate of 24.49% (Parham and Scott, 1980).

3.2.8 Ledford Island

The Ledford Island site is located at the head of Ledford Island on the Hiwassee River in southeastern Tennessee. Much of the site is today underwater as a result of the Chickamauga Dam project. The site is a Late Mississippian village with a plaza and a palisade, and occupation dates from AD 1400 to 1545 (Helms, 2012). The inhabitants were intensive maize agriculturalists. More than 400 burials were recovered, and approximately twenty percent of the burials were clustered in a cemetery in the northeast part of the village (Boyd and Boyd, 1991). The remaining burials were located throughout the village area, and 98% were primary interments while the remaining two percent were secondary burials. There is evidence to suggest that this site was more ceremonially significant than nearby contemporaneous sites, and there is clear evidence for status differentiation and community leaders (Helms, 2012). Demographic analysis demonstrated somewhat high numbers of infants and children, low numbers of juvenile and adolescents, and a particularly high number of adults (Boyd, 1986). Though it is possible that this is the result of differential preservation or biased aging techniques, Boyd (1986) believes that this is instead an accurate representation of the typical age-at-death for this population. This is supported by Helms's (2012:143) finding that the age at death distribution here is consistent with that of modern indigenous groups. The skeletal sample used in this analysis consisted of 343 individuals with an overall lesion rate of 20.41% (Helms, 2012).

3.2.9 Averbuch

Averbuch is a Mississippian site located on an upland slope nine kilometers north of the Cumberland River in north-central Tennessee. It consisted of a village of approximately eleven acres and contained three cemeteries (Eisenberg, 1991). Occupation has been dated between AD

1275 and 1400 and probably lasted only a short time, and was used intensively for only 25 to 50 years (Mick, 2011). The inhabitants were intensive maize agriculturalists and appear to have been ‘ordinary folk’ (not of particularly high status) and the site probably played a subordinate role within a larger regional network. Any within-site status differentiation appears to be based on gender and/or age. Males and females are equally represented in the skeletal sample, though infants are highly under-represented (Eisenberg, 1991). This is probably because rather than burying infants in the cemetery, they were usually buried near residential structures, and far fewer of these were excavated thoroughly (Berryman, 1981). The skeletal sample used in this analysis consisted of 732 individuals with an overall lesion rate of 39.07% (Eisenberg, 1991). The δ^{13}C isotopic sample consisted of four individuals with an average value of -8.00 (Schurr, 1992).

3.2.10 Cox

The Cox site is a Dallas phase Late Mississippian village and mound site located along the east bank of the Clinch River in northeastern Tennessee. It has been dated between approximately AD 1450 and 1650 (Harle, 2010). The inhabitants were intensive maize agriculturalists. The mound was built in three stages, but interments were only added in the third and final building stage. Excavation of the mound took place in the 1930s and excavation of the village in the 1960s (Vogel, 2007). Much of the later excavation was done by amateur archaeologists who took home many of the artifacts that they found, making status comparisons between individuals buried in the village and individuals buried in the mound impossible (Harle, 2010). However, biological analysis showed that there was no difference in health status between the village and mound interments, with a single exception: the occurrence of porotic hyperostosis, which was more common in the village than in the mound (Vogel, 2007). It is possible that this is a result of the presence of more children in the village and more men in the mound (Harle, 2010). Because of these caveats and the fact that all other indicators of health show no difference, I have once again combined the mound and the village samples for this analysis. The skeletal sample used in this analysis consisted of 190 individuals with an overall lesion rate of 27.37% (Vogel, 2007).

3.2.11 Hardin Village

Hardin Village sits on an alluvial terrace of the Ohio River in a valley in the Appalachian foothills in northeastern Kentucky. Occupation has been dated somewhere between AD 1500 and 1675 and the site is part of the Fort Ancient tradition. The site covers approximately 4.5 acres and is situated on rich bottomland. The inhabitants combined intensive maize agriculture with the hunting and gathering of wild resources (Cassidy, 1972). Occupation probably lasted about 120 years and the population at any one time was between 50 and 100 individuals (Broida, 1984). The site appears to have been abandoned before contact with Europeans. Infant and childhood mortality were high and females had a higher life expectancy than males for all ages (Cassidy, 1984). The skeletal sample used in this analysis consisted of 292 individuals with an overall lesion rate of 8.22% (Cassidy, 1972). The $\delta^{13}C$ isotopic sample consisted of 49 individuals with an average value of -11.64 (Broida, 1984).

3.2.12 Tinsley Hill

The Tinsley Hill site sits on the bluffs and alluvial plain of the east bank of the Cumberland River in southwest Kentucky. It consists of a village, a cemetery and a mound which are all associated with the Late Mississippian period (Lane, 1993). Occupation has been dated to two phases, lasting from approximately AD 1000 to 1100 and AD 1300 to 1500. All skeletal samples came from the cemetery (associated with the later phase, AD 1300 to 1500) and the mound structure contained no burials but merely served as a platform for a succession of elevated buildings during the earlier phase. The inhabitants were intensive maize agriculturalists, but riverine resources were also very important (Clay, 1997). Bioarchaeological analysis suggests that status at this site was not sharply differentiated (Lane, 1993). The skeletal sample used in this analysis consisted of 81 individuals with an overall lesion rate of 2.47% (Lane, 1993). The $\delta^{13}C$ isotopic sample consisted of 19 individuals with an average value of -8.60 (Schurr and Powell, 2005).

3.2.13 Norris Farms #36

The Norris Farms #36 site is located in the Illinois River Valley in west-central Illinois. The site consists of a cemetery and associated village (Stone and Stoneking, 1999). Occupation has been dated to AD 1300 and the site is affiliated with the Oneota complex, a notably different way of life than that of the preceding Mississippians who occupied the region. Social complexity appears to have been simpler than that of Mississippian societies (Milner et al., 1991). Subsistence was based on agriculture and the growing of maize, beans and squash, though hunting, fishing and the gathering of wild resources were still important. Most of the burials came from a large, low mound in the cemetery which was is use for perhaps a few decades (Milner et al., 1991). The age and sex distribution of the burials matches that expected from traditional societies, which is at least in part due to the chronic violence experienced by inhabitants of the site. This caused a higher proportion of young people to be included in the cemetery than would normally be represented (Milner et al., 1989). It appears that most of the people from the village were buried in this cemetery (Stone and Stoneking, 1999). The skeletal sample used in this analysis consisted of 170 individuals with an overall lesion rate of 44.71% (Milner et al., 1991). The $\delta^{13}C$ isotopic sample consisted of 5 individuals with an average value of -12.60 (Buikstra and Milner, 1991).

3.2.14 Lewis Creek Mound

The Lewis Creek Mound site is located on the southeast bank of Lewis Creek in north central Virginia. The mound is located on a broad alluvial floodplain near the center of the known burial mounds in interior Virginia (Gold, 2004). Bioarchaeological analysis suggests that the individuals interred here practiced a subsistence economy based on agriculture which included maize, though wild resources remained a substantial portion of the diet through time (Gold, 2004). Excavations by both amateur and professional archaeologists took place from 1920 onward, the majority of which were done by professionals in the 1960s. The mound had three distinct layers: the lowest contained primary inhumations in pits, the middle contained secondary burials as well as primary interments, and the top layer contained only secondary burials. Some graves contained artifacts like shell beads and stone and antler tools (Trimble, 1996). Radiocarbon dates on the bones place use of the mound between AD 1000 and 1160. The demographic profile of the well-excavated remains (37 individuals, from which the study sample used here was drawn) matches expectations for that of a complete population and contains fourteen adults, one young adult, and twenty-two infants and children. This may be a coincidence though, as the larger (though more problematic) looted sample of skeletons has many more adults than infants and children. Though the small sample size prohibits rigorous paleodemographic analysis, it appears at least that males and females of all ages were included in the mound (Gold, 2004). The skeletal sample used in this analysis consisted of 26 individuals with an overall lesion rate of 11.54% (Gold, 2004). The $\delta^{13}C$ isotopic sample consisted of 9 individuals with an average value of -12.38 (Trimble, 1996).

3.2.15 Juhle Ossuaries

The Juhle site is located on Nanjemoy Creek in southern Maryland. It is comprised of three ossuaries and a possibly associated habitation site. The individuals interred here were primarily agriculturalists who supplemented their diet with gathering and hunting of local wild resources (Chase, 1988). Ossuary I was excavated in the 1950s and Ossuary II was excavated in the 1970s, and both of these together provide the sample used in this analysis. Skeletal remains in each pit were found completely articulated, partially articulated, in bundles of partially disarticulated bones, or as scattered completely disarticulated bones (Ubelaker, 1974). Associated artifacts include shell beads and pottery (Chase, 1988). These indicate that Ossuaries I and II are nearly contemporaneous and were in use sometime before contact in the 16th century, between AD 1500 and 1600. The skeletal samples appear to represent the entire population fairly accurately, both with regard to age and sex (Ubelaker, 1974). The skeletal sample used in this analysis consisted of 208 individuals with an overall lesion rate of 17.79% (Chase, 1988).

3.2.16 Boytt's Field

The Boytt's Field site is located on a low rise in the midst of a broad field on the banks of the Ouachita River in south-central Arkansas. Excavations were carried out by amateur archaeologist Clarence Bloomfield Moore during a journey along the Ouachita River in 1908 and

1909. Burials were found both flexed and extended; some were complete and some were disarticulated (Moore and Hrdlička, 1909). The site was a part of the Plaquemine Mississippian (AD 1200-1650) culture wherein subsistence was based partly on maize agriculture and partly on wild resources (Rose et al., 1991). The skeletal sample used in this analysis consisted of 25 individuals with an overall lesion rate of 4.00% (Rose et al., 1991).

3.2.17 Ward Place

The Ward Place site was found and excavated by amateur archaeologist Clarence Bloomfield Moore during the same 1908-1909 trip during which the Boytt's Field site was found. It consists of a cemetery on a low rise of land in a broad field. All burials were extended and associated burial goods included stone and bone tools, ceramic items and shell beads (Moore and Hrdlička, 1909). Like Boytt's Field, the site was a part of the Plaquemine Mississippian (AD 1200-1650) culture and subsistence was based partly on maize agriculture and partly on wild resources (Rose et al., 1991). The skeletal sample used in this analysis consisted of 25 individuals with an overall lesion rate of 0.00% (Rose et al., 1991).

3.2.18 Buffalo

The Buffalo site is located on a high terrace of the Kanawha River, approximately 30 km east of where the Kanawha meets the Ohio River. The site has been attributed to the Fort Ancient tradition and is dated to AD 1500-1700 (Williams, 2005). The inhabitants were sedentary maize agriculturalists who supplemented their diet with locally hunted and gathered wild resources. There are at least two overlapping villages and occupation of these probably lasted for 30 to 50 years (Metress, 1971). The site itself consisted of a plaza, ceremonial structures and a palisade.

At least 562 burials were uncovered (Hanson, 1975), though only a portion of those were included in the Global History of Health database from which data in this project derive. Excavation took place between the 1930s and 1960s and was done both by amateurs and professionals (Metress, 1971). The skeletal sample used in this analysis consisted of 99 individuals with an overall lesion rate of 13.13% (Steckel et al., 2002).

3.2.19 Moundville

The Moundville site sits in the south bank of the Black Warrior River in west-central Alabama. Occupation lasted from the Late Woodland through the Mississippian, but the skeletal sample used here is restricted to the Mississippian and protohistoric periods (AD 1050-1700). These are all non-mound burials excavated by the Alabama Museum of Natural History in the 1930s and 1940s (Powell, 1991). The site expanded and grew more complex over time, and at its peak included more than twenty mounds, several multi-room structures, many small residential houses, and a palisade. The population at this time was probably around 3000 (Powell, 1991). Subsistence was based on maize agriculture with supplements from wild plants and animals. Social stratification was certainly present here, and it was likely based both on birth and achievement during life (Peebles and Kus, 1977). The skeletal sample is composed of approximately 25% subadults and 75% adults, and based on grave goods can potentially also be divided into approximately 14% elites, 34% sub-elites, and 52% non-elites (no grave goods at all) (Powell, 1991). The skeletal sample used in this analysis consisted of 162 individuals with an overall lesion rate of 9.26% (Powell, 1991). The $\delta^{13}C$ isotopic sample consisted of 37 individuals with an average value of -10.80 (Schoeninger and Schurr, 1998).

3.2.20 Eiden

The Eiden site is situated on a high bluff overlooking the French River in northern Ohio. This location allowed access to four distinct ecozones: an elm/ash swamp, mixed mesophytic forest, mixed oak forest, and prairie grasslands (Lallo and Blank, 1977). These provided resources like fish and deer, though the inhabitants of the site were full-time agriculturalists and maize was a substantial part of the diet. The site consists primarily of residential structures and the burials are scattered throughout the same area. It has been assigned to the Terminal Late Woodland period and has been radiocarbon dated to AD 1490 ± 55 (Lallo et al., 1977). The skeletal sample used here contained approximately 25% subadults and 75% adults, and the representation of each sex among the adults was approximately equal (Lallo and Blank, 1977). The skeletal sample used in this analysis consisted of 122 individuals with an overall lesion rate of 17.21% (Lallo and Blank, 1977).

3.2.21 Anderson

The Anderson site sits on the bank of the Little Miami River in southwestern Ohio. It includes habitation areas and a cemetery, and occupation has been dated from AD 1235 to 1400. The inhabitants were permanently sedentary maize agriculturalists who supplemented their diet with wild resources, and the site has been assigned to the Fort Ancient tradition (Lallo, 1979). Excavations were primarily done by amateur archaeologists in the late 19th and early 20th centuries (Griffin, 1966). Social stratification does not appear to have been a major component of Fort Ancient society (Griffin, 1992), and both adults and subadults were included in the cemetery. Those well-preserved enough to be included in Lallo's (1979) study were approximately 19% subadults and 81% adults. The skeletal sample used in this analysis consisted of 44 individuals with an overall lesion rate of 65.91% (Lallo, 1979). The $\delta^{13}C$ isotopic sample consisted of 8 individuals with an average value of -10.60 (Cook and Schurr, 2009).

3.2.22 East St. Louis Stone Quarry

The East St. Louis Stone Quarry site is a cemetery that sits on a low ridge which is part of the Falling Springs meander scar in southwest Illinois. The site consists of mortuary pits, postholes and fill, as well as limestone slabs that are part of a charnel structure that sits at the center of the cemetery (Milner, 1983). Radiocarbon dates place use between AD 1253 and 1295 (Emerson and Hargrave, 2000). Males, females and children were all represented and it appears to have been a non-elite cemetery. Inhumations were both primary and secondary, and the age and sex structure of the sample is comparable to that of other past populations (Milner, 1983). Stable isotope data indicates that diet included a mix of cultivated food like maize and local wild resources (Hedman et al., 2002). The skeletal sample used in this analysis consisted of 41 individuals with an overall lesion rate of 17.07% (Milner, 1991). The $\delta^{13}C$ isotopic sample consisted of 21 individuals with an average value of -10.96 (Hedman et al., 2002).

3.2.23 Kane Mounds

The Kane Mounds site consists of four low mounds spaced approximately 40 meters apart along a bluff overlooking the American Bottom region in southwest Illinois. Stable isotope data indicate that maize was a large part of the diet (Buikstra and Milner, 1991). Burials were placed in the mounds as well as under and around them, and the site is unusual in that it is unclear whether the mounds were man-made or a natural feature of the landscape. Radiocarbon dates of AD 1286 and 1293 have been produced for the site, though the site appears to have been in use over several generations, mostly during the Mississippian period (Emerson and Hargrave, 2000). Though involved in the Cahokian trade and exchange network, analysis of burial practices suggests that the ‘ethnic identity’ of those buried here was not necessarily Cahokian (Emerson and Hargrave, 2000). The skeletal sample used in this analysis consisted of 98 individuals with an overall lesion rate of 6.12% (Milner, 1991). The $\delta^{13}C$ isotopic sample consisted of 4 individuals with an average value of -10.30 (Buikstra and Milner, 1991).

3.2.24 Summary

As previously mentioned, there are some inconsistencies in this dataset which result from varying data collection strategies and questions of interest for the researchers who studied each of these sites. Demographic profiles are not the same or even known for each site, and the social status of individuals may vary both between and within sites. See Chapter 6 for a more in-depth discussion on the potential implications of these problems. A third inconsistency is that only some of the skeletal samples were definitively associated with a habitation area, while others were recovered from cemeteries. In the latter case, it is possible that the habitation areas were in a different location, making the environment at the cemetery irrelevant for the purposes of this study. However, the expansion of the geographic area from a single point to a 15 km radius around each site should mitigate this problem somewhat as the environmental data reflects averages over a broader area. This means that the ecological data reflect the environment of the general area in which people lived rather than the potentially anomalous characteristics of a particular small area.

Despite these differences, all of the people who lived at these sites were sedentary or semi-sedentary and relied primarily upon agriculture for subsistence. There were also 25 or more individuals buried at each site, and lesion rates varied between 0.00% and 65.91%. Though cultural practices varied regionally and the degree and nature of interactions between groups varied over time (Pollack et al., 2002; Blitz, 2010), the differences are not as extreme as they would be if the comparison being made was, for example, between Mississippian agriculturalists from the southeast and contemporary hunter-gatherers from the California coast. It is hoped that these sites are culturally similar enough that the effects of environment on health will be visible. Among other things, environmental differences can affect the kinds of parasites that individuals were likely to be exposed to, as discussed below.

3.3 Parasites

The parasites that I have considered in this analysis are *Necator americanus*, *Trichuris trichiura*, *Ascaris lumbricoides*, *Giardia lamblia*, *Diphyllobothrium* spp., and *Echinostoma* spp. As discussed below, each of these has a different life cycle during which they are affected by different environmental variables. They have also all been associated with anemia in clinical studies, and their presence in the pre-Columbian New World has been established through finds at sites other than those included in this analysis, as discussed below. Table 2 summarizes these parasites and their associated ecological variables.

3.3.1 Necator americanus (hookworm)

Necator americanus, also known as hookworm, is a soil transmitted helminth which is a member of the phylum Nematoda. It is one of the most common human parasite infections in the world, and according to the Centers for Disease Control and Prevention (CDC, 2014), infection is highest in places without adequate waste management facilities or where human feces are used

Table 2. Anemia-causing parasites and associated ecological variables.

Parasite	Ecological Variable	Expected Relationship	Source
Necator americanus (hookworm)	temperature	most prevalent 20-35°C; minimum 14°C	Brooker and Michael, 2000; Montenegro et al., 2006
	soil moisture	most prevalent in wetter soils, though complete saturation prevents embryonation	Brooker and Michael, 2000
	soil type	most prevalent in sandy, well-drained soils; less prevalent in clay soils	Anderson and Allen, 2011; Brooker and Michael, 2000
	altitude	most prevalent at lower altitudes	Brooker and Michael, 2000
Trichuris trichiura (whipworm)	temperature	most prevalent 32-35°C; minimum 5°C; maximum 45°C	Brooker et al., 2006; Brooker and Michael, 2000
	soil moisture	most prevalent in wetter soils	Brooker and Michael, 2000
	altitude	mixed findings	Brooker and Michael, 2000
Ascaris lumbricoides (giant roundworm)	temperature	most prevalent 28-32°C; maximum approx. 38-40°C	Brooker et al., 2006; Brooker and Michael, 2000
	soil moisture	most prevalent in wetter soils	Brooker and Michael, 2000
	altitude	mixed findings	Brooker and Michael, 2000
Echinostoma spp.	major lakes and rivers	most prevalent in areas with more major lakes and rivers	Hartson et al., 2011; Johnson and McKenzie, 2009
	water temperature	most prevalent in cooler water; optimum range 19-30°C	Evans, 1985
Diphyllobothrium spp. (fish tapeworm)	major lakes and rivers	most prevalent in areas with more major lakes and rivers	Scholz et al., 2009
	water temperature	most prevalent in cold water	Scholz et al., 2009
Giardia lamblia	temperature	mixed findings	Britton et al., 2010 (more prevalent at higher temperatures); Walsh, 2013 (more prevalent at lower temperatures)
	rainfall	more prevalent where there is greater rainfall	Britton et al., 2010; Cifuentes et al., 2004
	water temperature	most prevalent in cooler water	deRegnier et al., 1989

as fertilizer. Along with symptoms like diarrhea, vomiting and abdominal cramping, heavy infestations have been shown to cause anemia (Farid et al., 1969; Cox, 1993; Stoltzfus et al., 2004). This is particularly true for young children.

N. americanus is prevalent in tropical and subtropical areas of the modern world (Cox, 1993). Coprolites from the site of Pedra Furada, Piauí, Brazil demonstrate that hookworm was present in the New World as early as 5000 BC (Montenegro et al., 2006). There have also been finds from a number of pre-Columbian sites in the southern United States, including at Big Bone Cave in Tennessee, Upper Salts Cave in Kentucky, and Daws Island in South Carolina (Gonçalves et al., 2003). As is clear from the description of their life cycle below, an important portion of hookworm development takes place in the soil. This must take place under the correct environmental conditions, as outlined in Table 2. In particular, hookworm development has been shown to be affected by soil type and moisture, altitude, and temperature.

N. americanus life cycle:

- Eggs are deposited in the soil
- The eggs hatch into rhabditiform larvae after 1-2 days
- Over the course of 4-10 days the larvae molt twice and become infective filariform larvae
- These larvae enter a human body through the skin, and travel via the circulatory system into the heart and lungs
- The larvae move into the bronchial tree and into the throat where they are swallowed and move to the small intestine
- Here the larvae moult twice and develop into adult worms, attaching to the intestinal wall and resulting in blood loss; eggs are then passed into feces and the cycle begins again (Cox, 1993; CDC, 2014)

3.3.2 Ascaris lumbricoides (giant roundworm)

Ascaris lumbricoides, also known as the giant roundworm, is a soil transmitted helminth and the largest human intestinal nematode. It is one of the most prevalent human parasitic infections in the world (Cox, 1993). Infection is a result of ingesting infected eggs, usually via contaminated food. Immediate symptoms of infection are usually mild and can include abdominal cramping, and in some cases of heavy infestation, anemia has also been observed (Stoltzfus et al., 2004; CDC, 2014). Since part of the life cycle of this parasite takes place outside of its host, a number of environmental factors have been demonstrated to affect development. These are outlined in Table 2.

The modern distribution of *A. lumbricoides* is worldwide. The earliest physical evidence of its presence in the New World comes from the Huarmey Valley of Peru, where eggs were found in coprolites from 2277 BC (Gonçalves et al., 2003). However, aDNA analysis suggests that it may have been present as far back as 6850 BC in Brazil and Chile (Leles et al., 2008). In the southern United States, *A. lumbricoides* has been found at the site of Upper Salts Cave in Kentucky, Big Bone Cave in Tennessee, and Antelope House and Elden Pueblo in Arizona (Gonçalves et al., 2003). It has been observed that archaeological evidence for *A. lumbricoides* infection in the past is relatively uncommon in the New World when compared to the Old World, perhaps because of differential sanitation practices and/or access to anti-helminthic treatments (Leles et al., 2010; Reinhard et al., 2013). Nonetheless, the clear presence of *A. lumbricoides* in pre-Columbian North America, in combination with the high prevalence of the parasite today, makes it a strong possible cause of anemia in early agricultural populations in the eastern United States.

A. lumbricoides life cycle:

- Fertilized eggs are deposited in the soil
- The eggs embryonate and after 10-15 days the larvae moult inside the egg, becoming infective
- The infective eggs are swallowed and move to the small intestine where the stage 2 larvae hatch
- The larvae travel through the circulatory or lymphatic system to the heart and lungs where they moult twice
- The larvae move into the bronchial tree and into the throat where they are swallowed and move to the small intestine
- Here the larvae develop into adult worms; females can produce as many as 200,000 eggs per day which are then passed into feces and the cycle begins again

  (Cox, 1993; CDC, 2014)

3.3.3 Trichuris trichiura (whipworm)

Trichuris trichiura is a soil transmitted helminth that is also known as whipworm, so named for its long, thin posterior portion and wider anterior portion that resemble a whip (Cox, 1993). Infection is common in tropical regions of the world today, and also in temperate regions during warmer periods. Mild infections are usually symptomless, but heavy infections can lead to diarrhea, blood loss, and rectal prolapse. In children, growth stunting and cognitive impairment can also occur (CDC, 2014). Chronic infection can result in anemia in both children and adults (Farid et al., 1969; Cox, 1993; Carrilho Galvao et al., 2011).

The oldest finding of *T. trichiura* in the New World is eggs found in coprolites at the site of Lapa Pequena in Minas Gerais, Brazil. They have been dated to approximately 6050-5050 BC (Gonçalves et al., 2003). In North America, eggs have been found in latrine soils from the site of Elden Pueblo, Arizona, dated to AD 1070-1250 (Reinhard et al., 1987). Like *A. lumbricoides*, archaeological evidence for *T. trichiura* infection is less common in the New World than in the Old (Reinhard et al., 2013). However, given the imperfect nature of the archaeological record (preservation issues, variable laboratory and excavation methods, etc.), it is still highly possible that *T. trichiura* was an important cause of anemia in prehistoric North America.

T. trichiura life cycle:

- Eggs are deposited in the soil
- The eggs embryonate and after 15 to 30 days become infective
- The infective eggs are swallowed and move to the small intestine where the larvae hatch
- The larvae then move into the cecum where they mature into adults, attaching to the mucosa of the cecum and nearby areas of the large and small intestine; females can produce as many as 20,000 eggs per day which are then passed into feces and the cycle begins again

  (Cox, 1993; CDC, 2014)

3.3.4 Giardia lamblia

Giardia lamblia is a protozoan parasite that lives in the small intestine of humans and other animals. It is also known as *Giardia duodenalis*, *Giardia intestinalis* or *Lamblia intestinalis* (Cox, 1993). It is the most prevalent human intestinal parasite, and infection generally occurs as a result of eating contaminated food or water or from direct hand-to-mouth contact (Bogitsh et al., 2005). Symptoms of infection include diarrhea, abdominal cramping and nausea (CDC, 2014). It is associated with general malnutrition in many places, and specifically vitamin B_{12} deficiency (Olivares et al., 2002; Carvalho-Costa et al., 2007). Given the role that vitamin B_{12} deficiency may play in causing anemia, as well as the association between *G. lamblia* and anemic individuals in modern studies (Carrilho Galvao et al., 2011), it is certainly possible that it was an important cause of anemia in the past.

Today, *G. lamblia* is found in every part of the United States and all over the world (CDC, 2014). Its pre-Columbian presence in North America is evidenced by finds in coprolites from Big Bone Cave, Tennessee and Antelope House, Arizona (Gonçalves et al., 2003). These sites have been dated to 227 BC and AD 1200, respectively. As is clear from the life cycle outline below, part of the life of *G. lamblia* is spent outside the host. It is during this time that they are susceptible to environmental variables, as outlined in Table 2.

G. lamblia life cycle:

- Cysts with a rigid outer shell are deposited in water
- The infective cysts are swallowed and move to the small intestine where they excyst into trophozoites
- The trophozoites reproduce asexually and either attach to the walls or swim freely in the small intestine
- Some trophozoites encyst, and both cysts and trophozoites are passed into feces; only the cysts are infective, and if deposited in water the cycle begins again

(Bogitsh et al., 2005)

3.3.5 Diphyllobothrium spp. (fish tapeworm)

Diphyllobothrium is a genus of tapeworm which includes *Diphyllobothrium latum*, the largest tapeworm that can infect humans. *D. pacificum*, *D. cordatum*, and other species also infect humans, though less frequently (CDC, 2014). Infection is caused by the consumption of raw or undercooked fish, which serve as intermediate hosts for these parasites (Cox, 1993). Infection is sometimes asymptomatic, though abdominal discomfort, vomiting, diarrhea and weight loss can also occur. Vitamin B_{12} deficiency leading to anemia can also occur (Scholz et al., 2009; CDC, 2014).

The oldest *Diphyllobothrium* finds in the New World come from 10,000-4,000 year old mummies and coprolites from the coast of Peru (Reinhard, 1992). In North America, evidence of human infection in the past has been found at Buldir Island and Adak Island in Alaska (AD 1400-1700 and 1100 BC, respectively) and at the Schultz site in Michigan (350 BC-AD 450) (Le Bailly and Bouchet, 2013). The requirement that a portion of the life cycle be spent in the water and fish as intermediate hosts means that *Diphyllobothrium* require appropriate environmental conditions, as outlined in Table 2.

Diphyllobothrium life cycle:

- Eggs are deposited in water
- The eggs embryonate for 18-20 days
- The eggs hatch into coracidia, a developmental stage during which they can swim
- A crustacean eats the coracidia, and inside the crustacean they develop into proceroid larvae
- A small fish like a minnow eats the crustacean, and inside the fish the proceroid larvae develop into infective plerocercoid larvae

- The small fish is eaten by a larger fish, and the infective pleroceroid larva move into the muscle tissue of the larger fish
- These large fish are eaten by humans, and inside the small intestine the plerocercoid larva develop into adult tapeworms, attaching to the intestinal mucosa
- After 5-6 weeks, up to 1,000,000 immature eggs are released per worm per day which are then passed into feces and the cycle begins again

  (Cox, 1993; Bogitsh et al., 2005; CDC, 2014)

3.3.6 Echinostoma spp.

Echinostoma is a genus of trematode, which are intestinal flukes that infect humans through the consumption of raw or undercooked fish or molluscs. Specific species are difficult to tell apart, and nearly impossible to differentiate based on eggs alone (CDC, 2014). Symptoms of infection include vomiting, diarrhea, abdominal pain and fever (Sohn et al., 2011; CDC, 2014). Anemia has also be known to result from heavy infestations (Sohn et al., 2011).

The earliest find of *Echinostoma* in the New World comes from the site of Cueva de los Muertos Chiquitos in Durango, Mexico. Eggs were found in coprolites that date to approximately AD 500. They were found in association with other parasites, and the timing of this find when compared to other sites led the authors to conclude that these parasites probably moved into this area from areas further south in the New World (Jiménez et al., 2012). Though not as early as Cueva de los Muertos Chiquitos, the pre-Columbian presence of *Echniostoma* in the New World is also supported by their presence in a mummy from the site of Lapa do Boquete in Minas Gerais, Brazil, which has been dated to AD 750-1350 (Sianto et al., 2005). Like *Diphyllobothrium*, the requirement that a portion of the life cycle of *Echinostoma* be spent in the water and in intermediate hosts means that their development requires appropriate environmental conditions. These are outlined in Table 2.

Echinostoma life cycle:

- Eggs are deposited in water
- The eggs embryonate for 2-4 weeks
- The eggs hatch into miracidia, a developmental stage during which they can swim
- The miracidia then enter a mollusk, and inside the mollusk they develop into cercariae
- The cercariae either remain in the mollusk or enter the water and find a new host like a fish, tadpole, another mollusk even aquatic vegetation, and inside this new host the cercariae encyst and become infective
- This second host is eaten by humans, and inside the small intestine the cysts excyst and develop into adults
- These adults reproduce and eggs are passed into feces and the cycle begins again (Bogitsh et al., 2005)

3.3.7 Summary

Though the life-cycles of these parasites vary in the ways that they develop and infect humans, they have one important thing in common: each of these parasites requires a period of time outside their primary host (e.g. a human) in order to develop and become infectious. It is during this stage that environmental setting matters and conditions like temperature and rainfall must be optimal for the parasite to survive and develop. GIS can be used to collect and organize comparable ecological data, as discussed below.

3.4 GIS data

Environmental variables that have been shown to influence the geographic distributions of the parasites discussed above are listed in Table 2. Perhaps somewhat obviously, the climate today is not the same as it was in the past. However, broad differences between regions (e.g. it is colder in the north than in the south) are probably not drastically different today than they would have been in the past. Accepting this assumption, modern environmental data is useful for examining differences between sites in different ecological settings. For this analysis, I collected data on mean annual temperature, rainfall, soil type, altitude, and surface area of major lakes and rivers within a 15 km radius of each site. Soil moisture is not included on this list because it is correlated with rainfall and soil type, which I am already measuring. It is also correlated with vegetation cover (Brooker and Michael, 2000:262) which has certainly changed considerably with modernization. Water temperature is also not included, primarily because it is influenced by a host of factors that would not have affected early agricultural communities, including changes in aquatic biodiversity, runoff from factories and farms, and large-scale hydroengineering projects that provide water for irrigation and industry (Foley et al., 2005). Though water temperature is not always linearly related to air temperature because of factors like humidity and evaporation rate (Mohseni and Stefan, 1999; Erickson and Stefan, 2000), the two are still related and tend to cycle similarly over time (Pilgrim et al., 1998). Modeling water temperature data was deemed overly complex for this project, so I will rely upon air temperature and assume that exclusion of water temperature does not represent a drastic loss of data. More information on the sources of these data and how they were collected will be discussed in Chapter 4.

3.5 Historic data

As a further evaluation of the correlation between parasite infection and cribra orbitalia and porotic hyperostosis, I will also compare historic rates of hookworm infection with prehistoric rates of porotic hyperostosis and cribra orbitalia. This is possible thanks to Eric Thoman who kindly provided data from the Rockefeller Archive Center on historic hookworm infection in the southeastern United States (Thoman, 2009). This data was collected by the Rockefeller Sanitary Commission (RSC) from 1909 to 1915 and has the advantage of certainty as far as diagnosis of parasite infection. However, as with archaeological data, there are a number of caveats that should be kept in mind. In many cases the selection of patients studied was targeted or non-random, and record keeping was often inconsistent and varied from location to location (Thoman, 2009). Further, infection rates are on a county-level rather than based on a single location like an archaeological site. Finally, this historic data only considered hookworm infection and no data was collected by the RSC regarding other parasite infections.

4. METHODS

In order to look at the relationship between skeletal lesions (representing anemia) and environmental variables (representing parasite infection) it was necessary to gather and organize the skeletal and ecological data that were described in Chapter 3. First I will describe the criteria used to select skeletal samples and the ways in which lesion rates were calculated, and this will be followed by a discussion the methods used to collect environmental data using GIS. Next I will outline the statistical procedures used during analysis and describe how the final regression models were created. Finally I will outline my hypotheses regarding the relationship between the dependent and independent variables. All statistical analyses were conducted in SPSS 22.0 and all statistical hypothesis testing used a significance level (α) of 0.05.

4.1 Data gathering and entry procedures

The data on porotic hyperostosis and cribra orbitalia used in this analysis reflect the percentage of individuals with skeletal lesions (cribra orbitalia or porotic hyperostosis) at a given archaeological site. I have included only sites where data from 25 or more individuals are available. Though 30 is often cited as a minimum number for a ‘good’ sample, e.g. representative of the population under study (Hogg and Tanis, 2005), this number is somewhat arbitrary. Lowering my minimum to 25 instead allowed me to maximize the number of sites, resulting in a total of 22. I have not set a minimum for number of individuals for stable isotope data, since these studies often have much smaller sample sizes due to the destructive nature of the method. Sample sizes for the historic data were all well above 25. Nine of the 22 archaeological sites were located in counties where the RSC collected hookworm infection rates, and there are therefore nine sites with historic infection rates as well as prehistoric lesion rates.

Many published studies report rates of porotic hyperostosis and cribra orbitalia together since they are sometimes thought to have the same etiology. This is why the primary analysis considers the two lesions together, and why the combined lesion samples are the largest. However, the data from some sites, including those listed in the Global History of Health database (Steckel et al., 2002), are reported for each individual. To make the data from these sites as compatible as possible with other sources, an individual from the Global History of Health database was scored as having lesions present if either porotic hyperostosis or cribra orbitalia was observable and present. The lesions were scored as absent if both orbits or both parietals were observable and lesions were absent, or if only one orbit or parietal was observable and lesions were absent. Scoring of cribra orbitalia and porotic hyperostosis individually was simpler – cribra orbitalia was recorded as present or absent based on a single or both orbits depending on the skeletal material available, and porotic hyperostosis was recorded as present or absent based on a single or both parietals depending on the skeletal material available.

Age and sex groups were assigned by the original researchers. For the individuals recorded in the Global History of Health database (Steckel et al., 2002), sex was originally designated as either definitely male or female, probably male or female, uncertain because the individual was less than 15 years old, or undetermined. In this analysis I recorded both definite and probable females as female, and definite and probable males as male. Those who were uncertain because they were less than 15 years of age were recorded as children, and the rest of the individuals were recorded as adults. For skeletal data from other publications, the age designation (adult or child) and sex designation (male, female or unknown) determined by the original researcher were used here as well. By far the most common cutoff age separating adults and subadults was 15, though it ranges from age 10 at Toqua (Parham and Scott, 1980) to age 20 at Juhle (Chase, 1988).

I used GIS to obtain environmental data for each site. The best format for this kind of analysis is raster data, where information is represented by cells which are analogous to the pixels that make up digital pictures. Each cell has one value for a measurement or category, like average millimeters of annual rainfall or soil type (Zeiler, 1999). To get an average of a variable for each site, I found the modal or mean value for each cell within a 15 km radius of the approximate center of the site. This distance is intended to represent the maximum area that people potentially interacted with on a daily basis (Williams, 2005). The mean or modal value for each site was then entered into Excel for statistical analysis. The specific sources of data and the methods used for data processing for each environmental variable are described below. All data was collected in or transformed to use the World Geodetic System of 1984 (WGS 84) geographic coordinate system. See Appendix A for the maps created for each variable for each site.

4.1.1 Soil

Soil data was obtained from the Soil Survey Geographic (SSURGO) Database, produced and distributed by the soil survey staff of the Natural Resources Conservation Service (NRCS) of the United States Department of Agriculture (USDA). Soil data was downloaded for the hydrologic units surrounding each site. This data is available for download at http://www.arcgis.com/apps/OnePane/basicviewer/index.html?appid=a23eb436f6ec4ad6982000dbaddea5ea. Data were originally collected at a scale of 1:12,000 to 1:63,360. I used each of these vector files to create a raster file with 30 x 30 meter cells based on the dominant drainage class of each map unit. I then simplified the drainage categories by reclassifying the original drainage designations into simply "well drained" (originally well drained, moderately well drained, somewhat excessively drained, or excessively drained) and "poorly drained" (originally somewhat poorly drained, poorly drained, or very poorly drained). I restricted my maps to the cells within 15 km of the center of each site using the 'buffer' tool in ArcGIS, and then recorded the proportion of land with well-drained soil for each site.

4.1.2 Elevation

Elevation data were obtained from the Land Processes Distributed Active Archive Center (LP DAAC) of the United States Geological Survey (USGS). The dataset is called the Advanced Spaceborne Thermal Emission and Reflection Radiometer (ASTER) Global Digital Elevation Model (GDEM), Version 2. It can be downloaded at http://gdex.cr.usgs.gov/gdex/. This ASTER GDEM 2 data are distributed in raster format with a 1 arc-second grid (approximately 30 x 30 meters at the equator) and therefore no conversion of format was necessary. Elevation values are in meters above sea level and the vertical accuracy is between 10 and 25 meters. I restricted my maps to only the cells within 15 km of the center of each site using the 'buffer' tool in ArcGIS, and then recorded the average elevation over all cells for each site.

4.1.3 Temperature

Temperature data were obtained from the NRCS of the USDA, and were available as polygon maps derived from 30 arc-seconds Parameter-elevation Regressions on Independent Slopes Model (PRISM) raster grids. These original PRISM data were produced by the PRISM Climate Group of Oregon State University and can be downloaded at http://datagateway.nrcs.usda.gov. Since the original PRISM rasters contained 30 arc second cells, I used this same scale when converting the polygon maps to rasters. Temperature values are in degrees Celsius, and the value of each cell represents the average annual minimum temperature averaged over thirty years (1981-2010). I chose to use minimum rather than maximum temperature because the minimum survivable temperatures for parasites has been more consistently and firmly established than the maximum (Brooker and Michael, 2000). I restricted my maps to only the cells within 15 km of the center of each site using the ‘buffer’ tool in ArcGIS, and the value recorded for each site represents the average temperature for all cells within this buffer for each site.

4.1.4 Precipitation

Like temperature, precipitation data were obtained from the NRCS of the USDA, and were available as polygon maps derived from 30 arc-seconds Parameter-elevation Regressions on Independent Slopes Model (PRISM) raster grids. These original PRISM data were also produced by the PRISM Climate Group of Oregon State University. They can be downloaded at http://datagateway.nrcs.usda.gov. Since the original PRISM rasters contained 30 arc second cells, I again used this same scale when converting the polygon maps to rasters. Precipitation values are in millimeters, and the value of each cell represents the average annual rainfall averaged over thirty years (1981-2010). I restricted my maps to only the cells within 15 km of the center of each site using the ‘buffer’ tool in ArcGIS, and the value recorded for each site represents the average precipitation for all cells within this buffer for each site.

4.1.5 Surface area of major lakes and rivers

Data on the surface area of major bodies of water were obtained from the Multi-Resolution Land Characteristics (MRLC) Consortium, which is a partnership of United States Federal agencies led by the USGS. The data used are from the 2011 National Land Cover Database (NLCD) and can be downloaded at http://www.mrlc.gov/nlcd11_data.php. These NLCD data are Landsat satellite-based and are distributed in raster format with 30 x 30 meter resolution and therefore no conversion of format was necessary. Each cell was classified as one of eighteen possible landcover types (e.g. herbaceous wetlands, shrub/scrub, evergreen forest). After restricting my maps to only the cells within 15 km of the center of each site using the 'buffer' tool in ArcGIS, I recorded the number of cells designated "open water" within the buffer area. The value recorded for each site represents the proportion of land covered by open water for each site.

4.1.6. Summary of variables

- *Lesion frequency* – percentage of individuals at a site demonstrating either porotic hyperostosis, cribra orbitalia or both on any observable eye orbits or parietals, minimum sample size of 25
- *Porotic hyperostosis frequency* – percentage of individuals at a site demonstrating porotic hyperostosis on any observable parietal
- *Cribra orbitalia frequency* – percentage of individuals at a site demonstrating cribra orbitalia on any observable eye orbit
- *Age* – designated as child or adult, determined by original researcher and cutoff at most sites was age 15

- *Sex* – male or female, determined by original researcher and only available for adults
- *Historic infection rate* – percentage of individuals diagnosed with hookworm infection in a given county in the early 1900s
- $\delta^{13}C$ *average* – average bone collagen carbon isotope value, no minimum sample size
- *Soil drainage* – percentage of well-drained soil within a 15 km radius of a site
- *Elevation* – average meters above sea level within a 15 km radius of a site
- *Temperature* – average annual minimum temperature (°C) from 1981-2010 within a 15 km radius of a site
- *Precipitation* – average annual precipitation (mm) from 1981-2010 within a 15 km radius of a site
- *Surface area of major lakes and rivers* – percentage of land cover designated as open water within a 15 km radius of a site

4.2 Statistical procedures

I have treated each archaeological site as an individual case, looking at the group-level frequency of porotic hyperostosis and cribra orbitalia, environmental characteristics (as surrogate measures of parasite prevalence), and average stable isotope ratios where available. I used multiple linear regression for my primary analysis, with porotic hyperostosis and cribra orbitalia as dependent variables and environmental and isotopic values as independent variables. The relationship between lesion rates and collagen carbon isotope ratios was investigated both through simple linear regression and Spearman’s rank correlation. Finally, the relationship between historic and prehistoric data was investigated through both simple and multiple linear regression.

4.2.1 Linear regression

Simple linear regression, as used here to compare historic infection and prehistoric lesion rates, models the relationship between two variables by assessing the ability of the independent variable (here, historic infection rate) to predict the dependent variable (here, prehistoric lesion rate). This is done by creating a linear regression line that minimizes the sum of the squares of the difference between the observed data points and the regression line. Multiple linear regression works in much the same way, but utilizes multiple independent variables (here, environmental variables) to predict the dependent variable.

Simple linear regression can be thought of in two dimensions, having a slope (*b*) and an intercept (*a*). The regression line here takes on the form $y = a + bx$ where y is the dependent (outcome) variable and x is the independent (predictor) variable. Similarly, multiple linear regression is an extension of simple linear regression with more than one predictor variable. The equation for multiple linear regression is similar to that of simple linear regression and takes the form $predicted\ value = constant + B_1(variable1) + B_2(variable2)\ldots + B_N(variableN)$, where B_1 through B_N are partial regression coefficients for each variable.

Interpreting multiple linear regression results involves evaluating both the fit of the overall model and the explanatory power of each of the predictor variables. The predictive accuracy of the overall multiple linear regression model can be assessed by looking at the R^2 value, which quantifies the proportion of the variance in the dependent variable that can be 'explained' by the model. R-square ranges between 0 and 1, with a value of 1 meaning that the model can predict the outcome variable with 100% accuracy. The analysis of variance (ANOVA) F-test and associated p-value which tests the null hypothesis that there is no linear relationship between the dependent and independent variables.

After the overall model fit is assessed, the relative importance of each independent variable in the regression model can be examined. The most relevant values in this analysis will be the standardized regression coefficients, the partial correlation coefficients, and their associated t-statistics and p-values. The standardized regression coefficient tells us how important a variable is in the model since it tells how much the dependent variable changes with a one unit increase in that independent variable if the other independent variables are held constant. It is important to compare the contribution of multiple independent variables using the standardized rather than the unstandardized regression coefficients since the unit of measurement has an effect on the unstandardized coefficient.

Another way of expressing the relationship between the outcome variable and a particular predictor is through the partial correlation coefficient, which represents the correlation between the dependent and independent variable when the effects of the other variables are held constant. The partial correlation coefficient varies between -1 and 1, which represent perfect negative or positive association between the outcome variable and one of the predictor variables, holding all of the other predictors constant (Bush, 2012, p 129). For both the standardized regression and partial correlation coefficient, a positive value means that the dependent and independent variables have a positive relationship, and a negative value means that they have a negative relationship. Finally, the p-value tells us whether or not to reject the null hypothesis that there is no linear relationship between an individual independent variable and the dependent variable.

4.2.2 Assessment of subgroup homogeneity

Given the previously-discussed differences in anemia manifestation and vulnerability between males, females, and children, it is important to assess whether or not it is acceptable to combine these groups for analysis. Heterogeneity within a sample can sometimes obscure existing relationships or create the illusion of relationships that do not actually exist. It is for this reason that I performed a series of chi-squared tests of independence to compare lesion rates between these subgroups at each archaeological site which met the assumptions of the Pearson's chi-squared test (a minimum expected count of 5 for each cell). The null hypothesis for Pearson's chi-squared test is that lesion rate and age/sex are independent, and the alternative hypothesis is that they are related. The standardized residual value in each cell reflects how many standard deviations the observed count is above or below the expected count. I also ran a Pearson bivariate correlation test, which results in a correlation value between -1 and 1. A positive number means that the relationship between the variables is positive, and a negative number means that the relationship is negative. The closer the value is to -1 or 1, the stronger the relationship between the two variables.

These tests relate to two hypotheses with regard to lesion rates in these subgroups. The first is that children show higher lesion rates than adults. As previously discussed, RBC production centers in children take time to shift to the long bones, and children are also less likely to have built-up stores of vitamins and minerals in their bodies. Infants are particularly vulnerable when they are breastfeeding and their mother is deficient in iron or vitamin B_{12} since infants do not have previously built-up stores of this vitamin and mineral to draw upon. These factors mean that children are both more likely to acquire anemia when they experience vitamin and mineral deficiencies, and more likely to manifest this anemia as cranial lesions. The second hypothesis is that women demonstrate higher lesion rates than males. I predicted this would be true because when women breastfeed, they share the vitamins and minerals that they consume with their infant, and thus a parasite infection would deplete the body's stores much more quickly than in a male who does not have to share the vitamins and minerals he consumes with another body.

To test these hypotheses I ran two series of Pearson's chi-squared tests. The first series (see Table 3) examined males and females by lesion presence to explore the possibility of combining males and females into a single 'adult' category. Four sites met the minimum expected count requirement, and the p-values for all of these indicate that the null hypothesis, that sex and lesion presence are not related, should be retained. This means that males and females can indeed be combined for the purposes of this analysis. Further supporting this conclusion, bivariate correlation between male and female lesion rates (n = 12 sites) was statistically significant at the .001 level and resulted in a Pearson correlation value of 0.961, indicating a strong positive correlation.

Combining males and females into a single category, it was possible to expand the sample to include archaeological sites that reported lesion rates separated by age but not by sex. The results of chi-squared tests looking at the relationship between age and lesion presence are presented in Table 4. Nine sites met the minimum expected count requirement, and the p-value for five of these nine indicated that the null hypothesis should be rejected – that is, age and lesion presence are related. For four of the five sites where results were statistically significant the standardized residual was positive for children with lesions, indicating that the number of children with lesions was greater than expected. Though not true at all sites, the rejection of the null hypothesis at a majority of sites suggests that children and adults should be examined separately. Bivariate correlation of lesion rates for adults and children (n = 19 sites) gave a Pearson correlation value of 0.590, and though statistically significant at the .01 level, this is a weaker positive correlation than that seen for males and females. Further, at least some

Table 3. Chi-squared results for males and females by lesion presence.

Site		Female		Male		Pearson Chi-Square Value	p
		No	**Yes**	**No**	**Yes**		
Anderson	Observed Count	5	12	9	10	1.217	.270
	Expected Count	6.6	10.4	7.4	11.6		
	Standard Residual	-0.6	0.5	0.6	-0.5		
Averbuch	Observed Count	113	88	118	104	.400	.527
	Expected Count	109.8	91.2	121.2	100.8		
	Standard Residual	0.3	-0.3	-0.3	0.3		
Cox	Observed Count	46	16	56	13	.919	.338
	Expected Count	48.3	13.7	53.7	15.3		
	Standard Residual	-0.3	0.6	0.3	-0.6		
Toqua	Observed Count	53	16	68	14	.880	.348
	Expected Count	55.3	13.7	65.7	16.3		
	Standard Residual	-0.3	0.6	0.3	-0.6		

Table 4. Chi-squared results for adults and children by lesion presence.

Site		Adult		Child		Pearson Chi-Square Value	p
		No	**Yes**	**No**	**Yes**		
Monongahela	Observed Count	60	6	29	26	22.484	.000
	Expected Count	48.5	17.5	40.5	14.5		
	Standard Residual	1.6	-2.7	-1.8	3.0		
Buffalo	Observed Count	47	7	40	5	.079	.779
	Expected Count	47.5	6.5	39.5	5.5		
	Standard Residual	-0.1	0.2	0.1	-0.2		
Sunwatch	Observed Count	46	9	58	16	.558	.455
	Expected Count	44.3	10.7	59.7	14.3		
	Standard Residual	0.2	-0.5	-0.2	0.4		
Averbuch	Observed Count	262	197	184	89	7.657	.006
	Expected Count	279.7	179.3	166.3	106.7		
	Standard Residual	-1.1	1.3	1.4	-1.7		
Hardin Village	Observed Count	130	10	138	14	.413	.520
	Expected Count	128.5	11.5	139.5	12.5		
	Standard Residual	0.1	-0.4	-0.1	0.4		
Eiden	Observed Count	86	5	15	16	34.512	.000
	Expected Count	75.3	15.7	25.7	5.3		
	Standard Residual	1.2	-2.7	-2.1	4.6		
Cox	Observed Count	102	29	36	23	5.807	.016
	Expected Count	95.1	35.9	42.9	16.1		
	Standard Residual	0.7	-1.1	-1.0	1.7		
Toqua	Observed Count	121	30	75	36	5.359	.021
	Expected Count	113	38	83	28		
	Standard Residual	0.8	-1.3	-0.9	1.5		
Juhle	Observed Count	134	10	37	27	37.632	.000
	Expected Count	118.4	25.6	52.6	11.4		
	Standard Residual	1.4	-3.1	-2.2	4.6		
Ledford Island	Observed Count	165	49	108	21	2.170	.141
	Expected Count	170.3	43.7	102.7	26.3		
	Standard Residual	-0.4	0.8	0.5	-1.0		

correlation would be expected between adults and children if anemia is indeed the result of parasitic infection, since the adults and children in question live in the same environmental setting.

In summary, the results of chi-squared and bivariate correlation tests suggest that adult males and females can be combined into a single ‘adult’ category, while children and adults should be separated for regression analysis. This is intended to help prevent heterogeneous subgroups from obscuring the relationships between lesion rates, environment and diet. The next step was to create the multiple linear regression model to be used for analysis.

4.2.3 Creating a regression model

Linear regression relies on a number of assumptions. For simple linear regression, these include: (1) that observations are independent; (2) that the relationship between the dependent and independent variables is linear; (3) for each value of the independent variable, the dependent variable is normally distributed; and (4) these distributions of the dependent variable have the same variance. Multiple linear regression has the same assumptions, though assumption (3) above changes slightly so that for each combination of independent variables (rather than each individual value) it is assumed at the dependent variable is normally distributed (Norušis, 2012).

The linearity of the relationship between variables can be verified prior to the analysis by examining scatterplots, and transformations can often be used to help make relationships more linear. Outliers for the dependent variable can be identified through examination of standard deviations – though what constitutes an ‘outlier’ has many definitions (Gray and Kinnear, 2012; Norušis, 2012; Harris and Jarvis, 2014), a commonly used rule of thumb is that any value of the dependent variable that is more than two standard deviations above the mean is an outlier (Anderson et al., 2014). Outliers are usually removed because these cases can have a disproportionate influence on results. After the analysis has been performed, examination of the residuals can provide further information as to whether or not the assumptions of linear regression have been violated. These include looking at studentized deleted residuals, which communicate how well a model would be able to predict a case if that case were not used to build the model. High values can indicate disproportionate influence on the model, and if the assumptions of linear regression are met, these values should follow a t-distribution. A t-distribution calculator (http://surfstat.anu.edu.au/surfstat-home/tables/t.php) was used with the appropriate degrees of freedom and an α of 0.05 to find the cutoff t values for identifying outliers (Anderson et al., 2014, p 660). Partial regression plots were also examined, and collinearity tolerance values all had to be above 0.1 (a minimum suggested by Norušis, 2012, p 540).

These diagnostic criteria were examined for each of the models created in this analysis, and any changes to the samples that were made as a result are discussed along with the results of each statistical test.

4.3 Statistical hypotheses

As previously discussed, children and adults were analyzed separately since their lesion rates were significantly different. I assessed the influence of maize-intensive diet on lesion rates by looking at carbon stable isotope values, and looked at porotic hyperostosis and cribra orbitalia separately for each age group. I also compared historic rates of hookworm infection with prehistoric lesion rates. The raw data used for these analyses can be seen in Appendix B. Below I outline how the general hypotheses discussed in section 2.8 will be supported or refuted statistically. The hypothesis that women would correlate better than men is not discussed since the results of chi-squared analysis here suggested that males and females should be combined.

Table 5 summarizes the expected direction of the relationship between each environmental variable and lesion rates.

1) *Hypothesis: there will be significant correlation between prehistoric lesion rates and environmental variables*

This hypothesis will be tested through multiple linear regression. The expected direction of relationships between the prevalence of particular parasites and ecological variables are listed in Table 5. The more often expectations are met for particular ecological variables, the stronger the support for the presence of the associated parasites. In some cases, previous studies found mixed results with regard to the nature of the relationship between a parasite and an ecological variable. Table 5 does not incorporate those mixed findings into the hypotheses laid out. This means that if results of this analysis demonstrate a relationship that is the opposite of that expected for a particular variable, it does not necessarily imply that a particular parasite did not contribute to anemia. If the expectations in Table 5 are not met, it is possible instead that the environmental requirements for a particular parasite to survive in the southeastern United States are not the same as those for a parasite living in the locations of previous studies.

The expected direction of association is generally in agreement when multiple parasites are associated with the same ecological variable, but there is one exception. The variable where there is disagreement, based on previous studies, is temperature. This is the result of using mean land surface temperature as a proxy for water temperature. An association between high frequencies of anemia and cooler temperatures could support the hypothesis that the three parasites associated with water (*Echinostoma* spp., *Diphyllobothrium* spp. and *G. lamblia*) were

Table 5. Ecological variables and their expected relationships with PH and CO.

Ecological Variable	Parasites Affected	Expected Direction of Association for Parasites	Overall Expected Direction of Association
temperature	*Necator americanus* *Trichuris trichiura* *Ascaris lumbricoides*	most prevalent 20-35°C; minimum 15°C most prevalent 32-35°C; minimum 5°C; maximum 45°C most prevalent 28-32°C; maximum approx. 38-40°C	positive correlation (since the average temperature at no sites exceeded the maximum tolerable temperature)
(water temperature)	*Giardia lamblia* *Echinostoma* spp. *Diphyllobothrium* spp.	most prevalent in cold water most prevalent in cooler water; optimum range 19-30°C most prevalent in cold water	negative correlation
rainfall	*Giardia lamblia*	more prevalent where there is greater rainfall	positive correlation
(soil moisture)	*Necator americanus* *Trichuris trichiura* *Ascaris lumbricoides*	most prevalent in wetter soils, though complete saturation prevents embryonation most prevalent in wetter soils most prevalent in wetter soils	positive correlation
soil type	*Necator americanus*	most prevalent in sandy, well-drained soils; less prevalent in clay soils	positive correlation
altitude	*Necator americanus* *Trichuris trichiura* *Ascaris lumbricoides*	most prevalent at lower altitudes mixed findings mixed findings	negative correlation
major lakes and rivers (surface area)	*Echinostoma* spp. *Diphyllobothrium* spp.	most prevalent in areas with more major lakes and rivers most prevalent in areas with more major lakes and rivers	positive correlation

causal factors of anemia, while an association with warmer temperatures could implicate the three soil transmitted helminths (*N. americanus*, *T. trichiura*, and *A. lumbricoides*).

Statistically, the general hypothesis that parasite infection was a cause of anemia will be supported if the environmental variables (serving as a proxy for parasite infection) can predict rates of porotic hyperostosis and cribra orbitalia (representing anemia) for both adults and children. I therefore predict that the R^2 values for both groups will be statistically significant, though I do not expect that they will be 1 given the many potential etiologies for anemia. The direction of association between the dependent variable and independent variables is reported as the standardized regression coefficient (β) which is either positive or negative. I predict that where the expected direction of correlation in Table 5 is positive that β will be positive, and where the expected direction of correlation is negative, β will be negative.

2) *Hypothesis: children will show stronger correlation than adults*

This will be tested through multiple linear regression and comparing the results for children and adults. If true, this will be supported statistically by higher R^2 values for children than for adults. It is also possible that the findings for children will be more in agreement with expectations with regard to relationship strength and direction based on the predictions in Table 5.

3) *Hypothesis: cribra orbitalia and porotic hyperostosis will correlate better separately than when grouped together*

Unfortunately, sample sizes are too small for rigorous statistical testing of this hypothesis, but to explore these relationships, I will compare the results of multiple linear regression looking at each lesion separately for children and adults. This hypothesis will be supported if the R^2 values of regression for both age groups for the individual lesions are higher than the R^2 values obtained when the lesions were combined. Further, the directions of relationships between lesions and environmental variables may meet expectations better when the lesions are separated.

4) *Hypothesis: $\delta^{13}C$ values will be able to successfully predict prehistoric lesion rates*

I will test this through simple linear regression and Spearman's rank correlation for both adults and children. The latter test procedure replaces the data values with ranks and tests whether or not there is a monotonic relationship and can be useful when there is a relationship between two variables but it is not linear. Correlation will be confirmed by a statistically significant p-value for either.

5) *Hypothesis: historic rates of infection and prehistoric lesion rates will be similar*

This will be tested using simple linear regression to see if historic infection rates can predict prehistoric lesion rates. Though the historic data is not reported separately for adults and children, these rates can still serve as a proxy for hookworm density. If this hypothesis is true then it will be statistically supported by a p-value that is statistically significant.

I will also test whether or not the environmental variables can successfully predict historic infection rates using multiple linear regression, and this would be confirmed by a high R^2 value and a p-value indicating statistical significance. Further, I predict that soil drainage will contribute the most to the model, since Anderson and Allen (2011) found correlation with soil types in their study using data from the same historic records. This will be supported if the standardized coefficient and partial correlation values for soil drainage is larger than those of the other variables included in the model.

5. RESULTS

This section will describe the statistical results obtained for each group analyzed. As previously mentioned, some subgroups such as children with porotic hyperostosis have smaller sample sizes because not every investigator reported skeletal lesion rates in such a way that porotic hyperostosis and cribra orbitalia could be separated. These smaller sample sizes often yielded results that were statistically insignificant; however, since it is more difficult to detect significance in smaller sample sizes, the patterns in the data are nonetheless discussed in Chapter 6.

5.1 Regression for adults

A square root transformation was used to improve linearity between adult lesion rate and the environmental variables. Though the relationship with water still does not appear to be particularly linear (see Figure 3), the two sites that ‘drive’ the relationship are Eiden and Juhle, and both of these are located on the edge of a major body of water (Lake Erie and the Atlantic Ocean, respectively). It may in fact be that water is only an important variable when the body of water nearby is very large, and since this could be a meaningful relationship, I have retained water as a variable in the model.

Two sites were removed from the sample as outliers. Anderson was removed since the lesion rate for this site was 2.8 standard deviations above the mean, and Averbuch had an unusually high studentized deleted residual (2.74). Using 14 degrees of freedom and an α of 0.05, the t distribution suggests that any studentized deleted residual value greater than 2.145 or less than -2.145 is an outlier (Anderson et al., 2014, p 660). Since Averbuch meets this criterion, is was removed from this analysis.

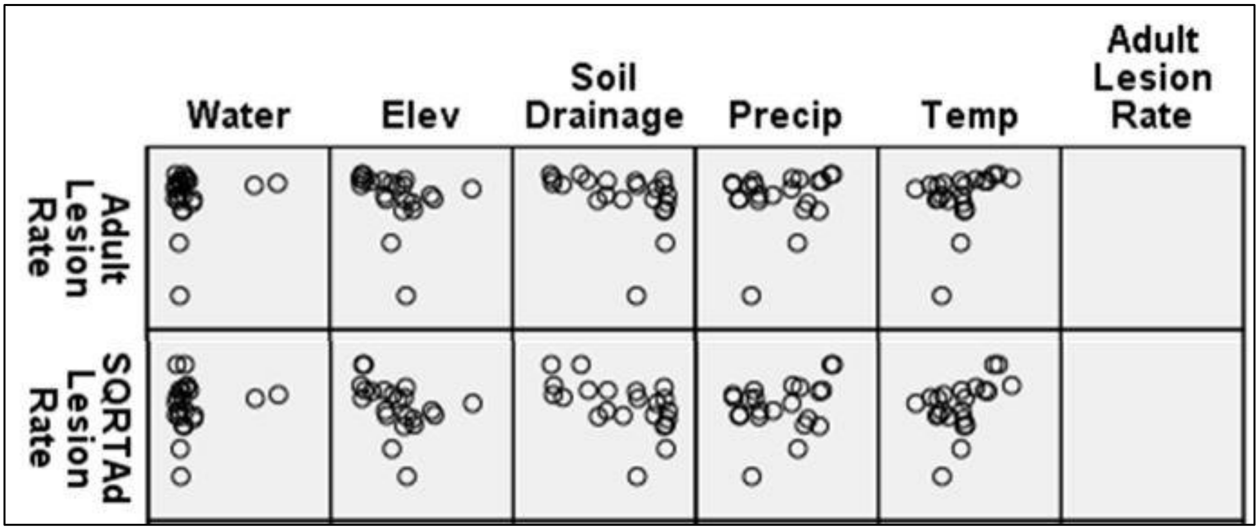

Figure 3. Scatterplots of relationships between adult lesion rate and environmental variables.

Table 6 contains the result of multiple linear regression for adults. The R^2 value demonstrates that 53.2% of the variability in adult lesion rates can be explained by the five ecological variables, and these results are statistically significant ($p = 0.040$). The standardized coefficient and partial correlation values indicate that soil drainage contributes most to the model, and the p-value for this variable is the only one that is statistically significant ($p = 0.045$). Soil drainage is followed in importance within the model by precipitation and then elevation. Temperature and water surface area contribute the least. The relationship with elevation and soil drainage is positive, and the relationship with temperature, precipitation and water surface area is negative. Figure 4 plots the observed values against the predicted values for the dependent variable, and the relatively even distribution of points on either side of the line suggests that model fit is good – however, it appears that the model may be under-predicting for lower values of the observed adult lesion rates.

Table 6. Results of multiple linear regression with transformed adult lesion rates.

R^2	Standard Error of the Estimate	p
.532	. 10855	.040

	Unstandardized Coefficient	Standardized Coefficient	t	p	Partial Correlations	Collinearity Tolerance
Constant	.448		.927	.370		
Soil Drainage	.205	.512	2.196	.045	.506	.614
Elevation	.000	.169	.439	.668	.116	.225
Temperature	-.001	-.042	-.087	.932	-.023	.146
Precipitation	-.006	-.297	-.879	.394	-.229	.294
Water	-.011	-.008	-.040	.969	-.011	.744

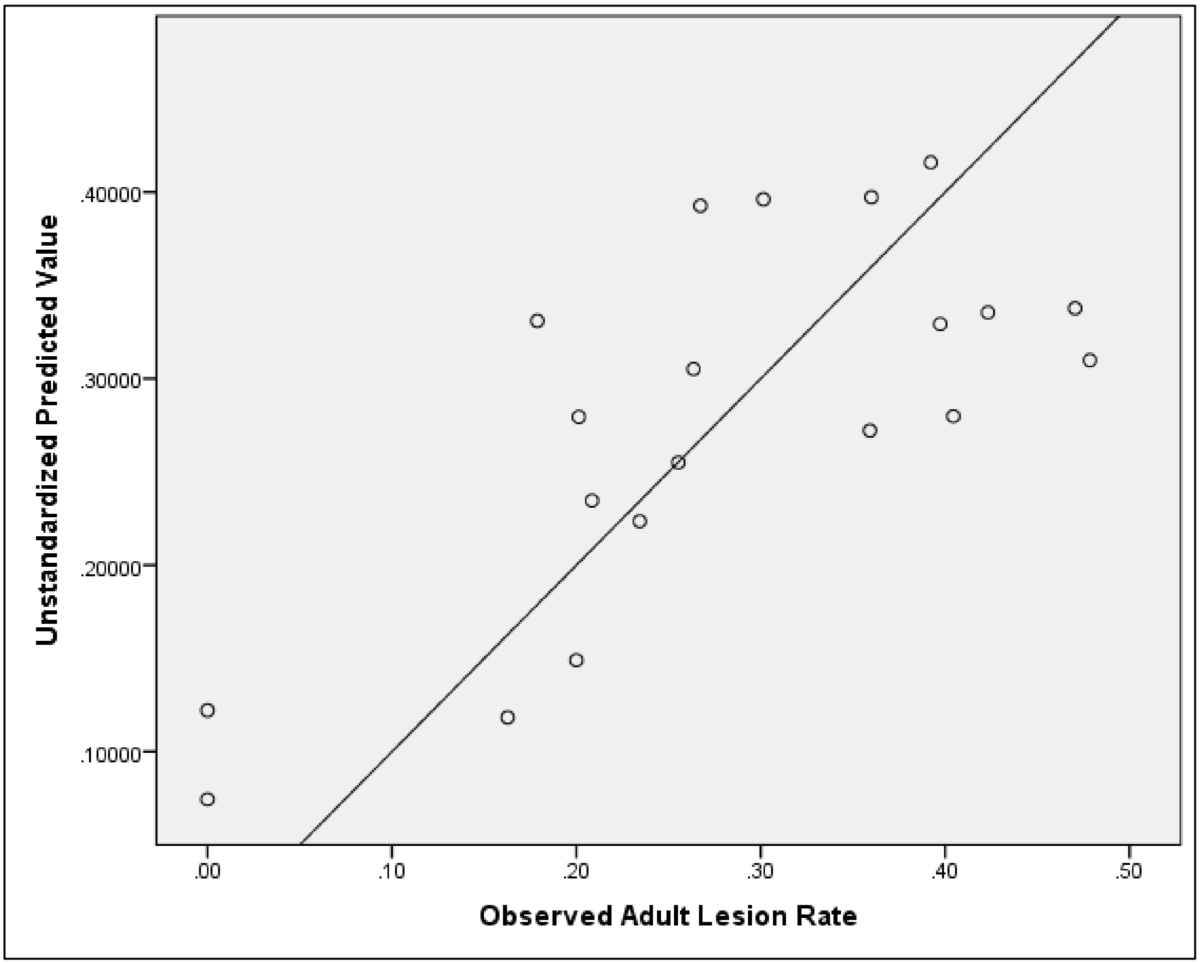

Figure 4. Plot of predicted adult lesion rate against the observed.

5.2 Regression for children

As with adults, a square root transformation was used to improve linearity between child lesion rate and the environmental variables. Though the relationship with water again does not appear linear (see Figure 5), I have left it as part of the model again and for the same reason, since it is the two same sites that 'drive' the relationship. Using 12 degrees of freedom and an α of 0.05, the t distribution suggests that any studentized deleted residual value greater than 2.179 or less than -2.179 is an outlier (Anderson et al., 2014, p 660). Anderson met this criterion (studentized deleted residual = 2.29) and was therefore removed from the sample. When Anderson was removed from the sample and the residuals reanalyzed, the studentized deleted residual for Norris Farms #36 had a value of 3.10 and so was also removed as an outlier.

The results of multiple linear regression for children can be seen in Table 7. The R^2 value suggests that the environmental variables can explain 63.7% of the variability in children's lesion rates, and the results are statistically significant ($p = 0.029$). The standardized coefficient and partial correlation values indicate that surface area of water contributes most to the model, and this is the only variable with a statistically significant relationship to child lesion rate ($p = 0.004$). The next most important contributors are soil drainage and then elevation. Temperature and precipitation contribute the least. The relationship between the dependent variable and each of the independent variables is positive. Figure 6 plots the observed child lesion rates against those predicted by the model, and the even distribution of points around the line suggests that model fit is good, though once again, the model may be under-predicting somewhat for lower values of the observed lesion rates.

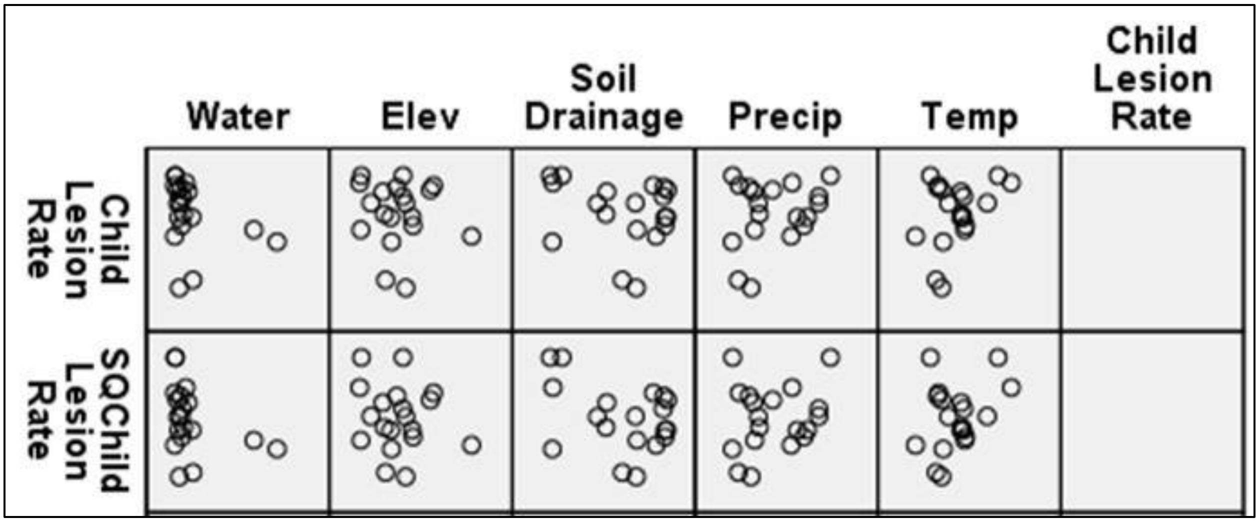

Figure 5. Scatterplots of relationships between child lesion rate and environmental variables.

Table 7. Results of multiple linear regression for children, five environmental variables.

N	R^2	Standard Error of the Estimate	p
17	.637	.15761	.029

	Unstandardized Coefficient	Standardized Coefficient	t	p	Partial Correlations	Collinearity Tolerance
Constant	-.585		-.676	.513		
Soil Drainage	.274	.450	1.931	.080	.503	.607
Elevation	.001	.413	.990	.343	.286	.190
Temperature	.008	.185	.365	.722	.110	.129
Precipitation	.005	.139	.419	.683	.125	.299
Water	1.530	.795	3.694	.004	.744	.712

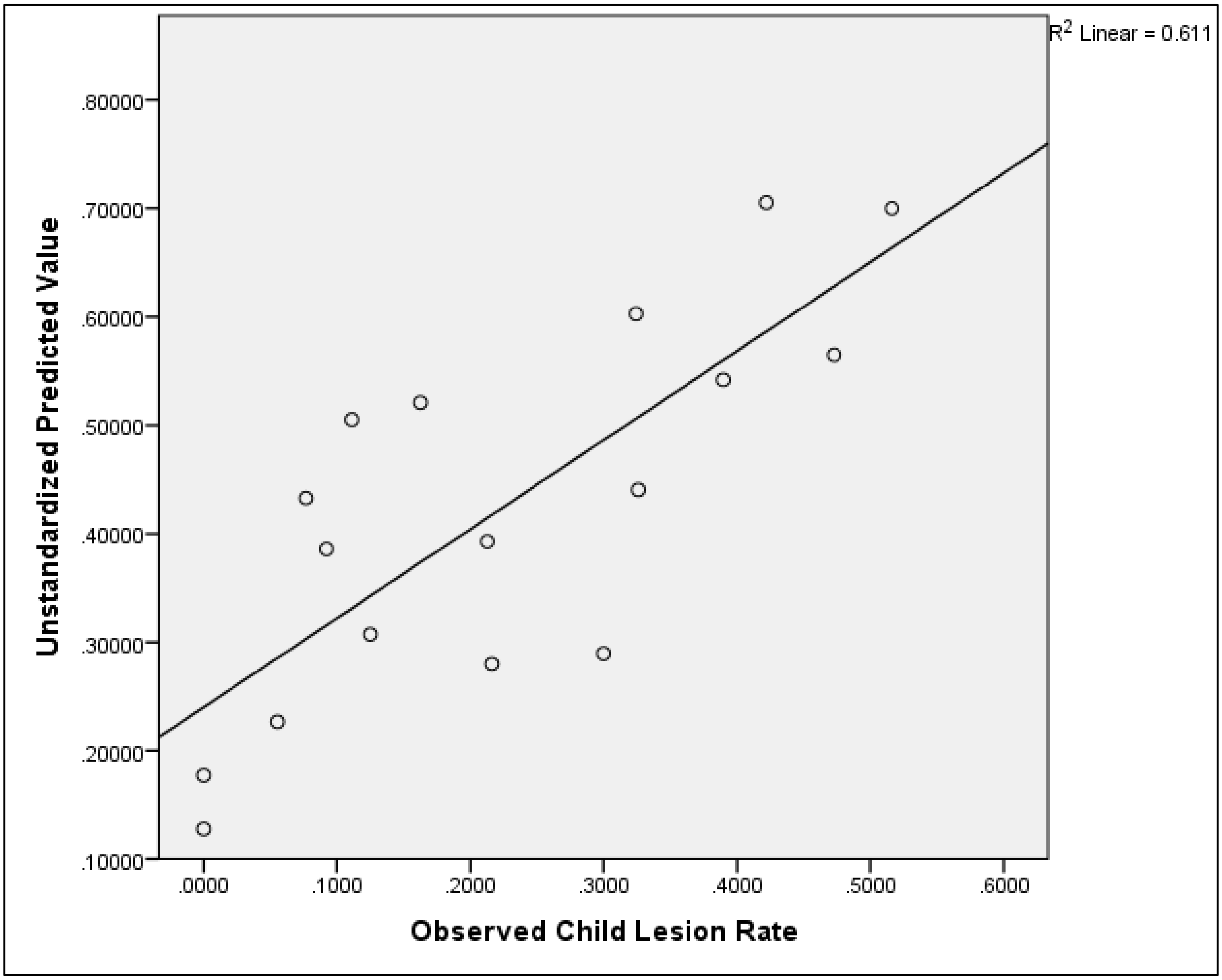

Figure 6. Plot of predicted child lesion rate against the observed.

5.3 Regressions by lesion type

As previously discussed, these analyses were done as an exploratory maneuver rather than to draw any statistical conclusions since sample sizes were very small (*n* = 8 sites) for each analysis. The following results do not include elevation as an independent variable because multicollinearity was an issue. No null hypothesis statistical tests were conducted, so p-values are not reported here. Given the exploratory nature of each of these analyses, no cases were removed as outliers and no transformations were applied.

5.3.1 Regression for porotic hyperostosis in children

The regression results for porotic hyperostosis rates in children can be seen in Table 8. Figure 7 plots the predicted porotic hyperostosis rate against the observed one, and model fit looks acceptable though it is difficult to say with much certainty given the small sample size. The analysis yielded an R^2 of 0.887, and temperature, precipitation and surface area of water all contributed significantly to the model. Soil drainage contributed the least. The relationships with soil drainage and temperature were negative, while the relationships with precipitation and surface area of water were positive.

5.3.2 Regression for cribra orbitalia in children

Regression results for cribra orbitalia rates in children can be seen in Table 9. The model resulted in an R^2 value of 0.661, and the standardized coefficient and partial correlation values demonstrate that precipitation and surface area of water contribute the most to the model, followed by temperature and then soil drainage. The relationship between cribra orbitalia rate and temperature is negative, while the relationship with the other three independent variables is positive. Figure 8 plots the observed and predicted cribra orbitalia rates in children and model fit appears satisfactory, though as before model fit is difficult to assess with such a small sample size.

Table 8. Results of multiple linear regression for porotic hyperostosis in children.

N	R^2	Standard Error of the Estimate
8	.887	.0618098

	Unstandardized Coefficient	Standardized Coefficient	t	p	Partial Correlations	Collinearity Tolerance
Constant	.163		.665	.554		
Soil Drainage	-.082	-.248	-.906	.432	-.464	.503
Temperature	-.022	-1.034	-3.332	.045	-.887	.393
Precipitation	.021	.883	3.250	.047	.882	.512
Water	1.008	.885	3.768	.033	.909	.685

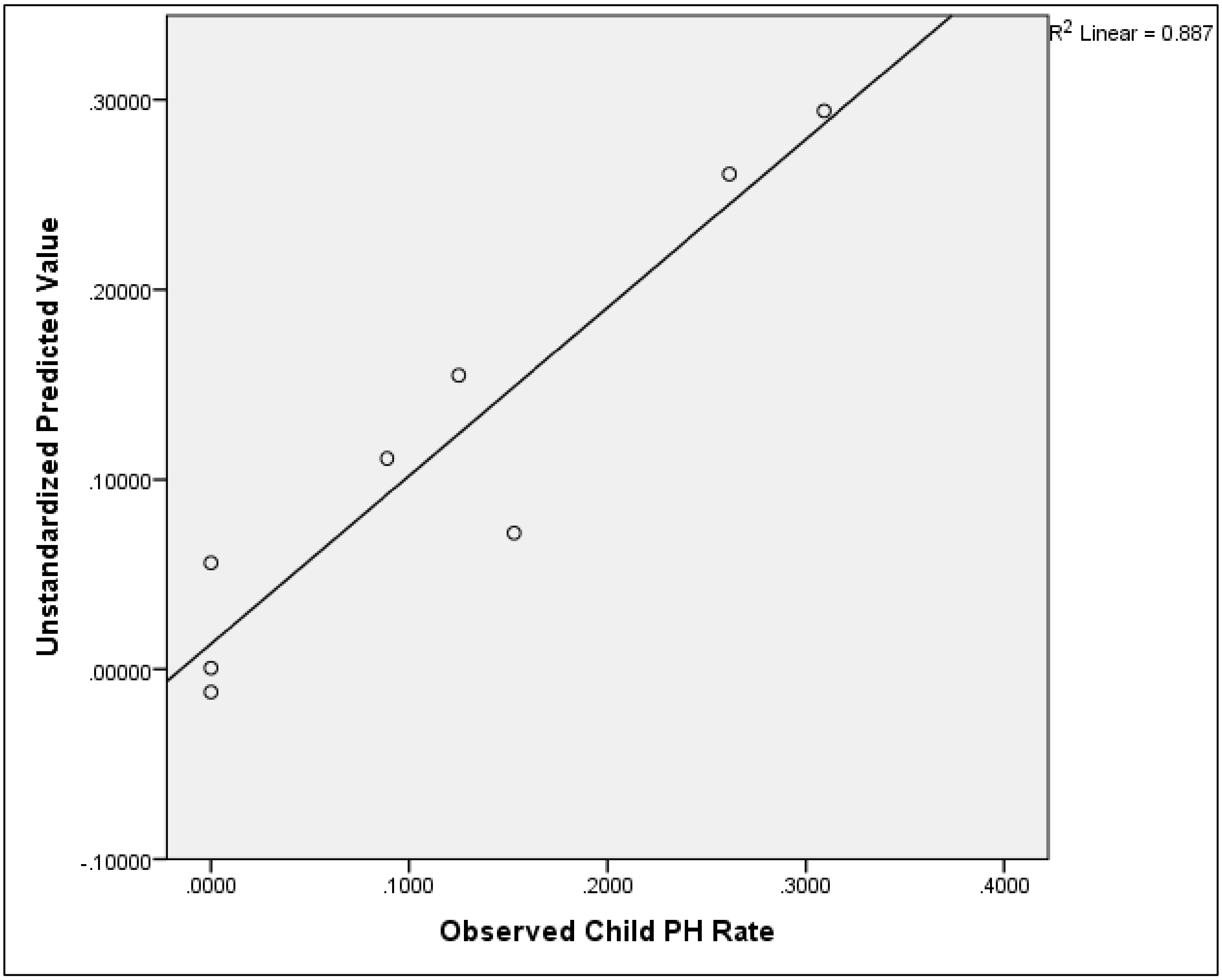

Figure 7. Plot of predicted child porotic hyperostosis rate against the observed.

Table 9. Results of multiple linear regression for cribra orbitalia in children.

N	R^2	Standard Error of the Estimate
8	.661	.1656863

	Unstandardized Coefficient	Standardized Coefficient	t	p	Partial Correlations	Collinearity Tolerance
Constant	-.597		-.907	.431		
Soil Drainage	.102	.200	.423	.701	.237	.503
Temperature	-.008	-.253	-.472	.669	-.263	.393
Precipitation	.023	.638	1.357	.268	.617	.512
Water	1.014	.574	1.414	.252	.632	.685

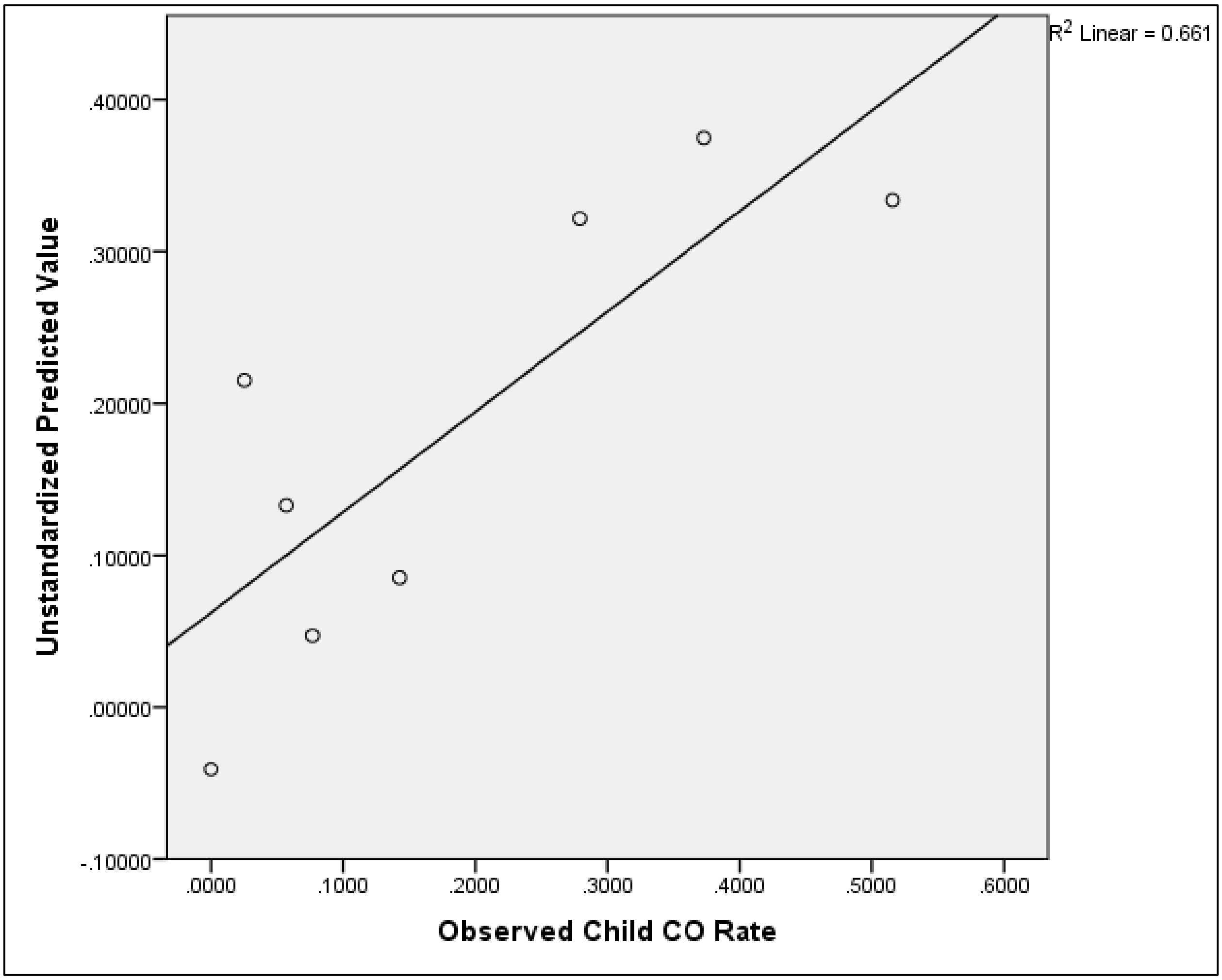

Figure 8. Plot of predicted child cribra orbitalia rate against the observed.

5.3.3 Regression for porotic hyperostosis in adults

Analysis of porotic hyperostosis in adults gave the results seen in Table 10. Figure 9 plots the predicted and observed porotic hyperostosis rates against one another – model fit appears to be acceptable, though the small sample size makes this conclusion uncertain. Regression resulted in a model with an R^2 value of 0.679. The standardized coefficient and partial correlation values demonstrate that soil drainage contributed the most to the model, followed by surface area of water and then precipitation. Temperature contributed the least to the model. Precipitation and surface area of water had a negative relationship with porotic hyperostosis rate in adults, and soil drainage and temperature had a positive one.

5.3.4 Regression for cribra orbitalia in adults

The results of regression for cribra orbitalia in adults can be seen in Table 11. The R^2 value is 0.727. The standardized coefficients and partial correlations demonstrate that precipitation contributes the most to the model, followed by water, temperature and then soil drainage. All relationships but temperature are positive. Figure 10 shows the predicted cribra orbitalia rates plotted against the observed, and the model fit looks adequate, though it is again difficult to assess with a small sample.

Table 10. Results of multiple linear regression for porotic hyperostosis in adults.

N	**R^2**	**Standard Error of the Estimate**
8	.679	.0534183

	Unstandardized Coefficient	**Standardized Coefficient**	**t**	**p**	**Partial Correlations**	**Collinearity Tolerance**
Constant	.145		.683	.544		
Soil Drainage	.149	.880	1.910	.152	.741	.503
Temperature	.005	.421	.806	.479	.422	.393
Precipitation	-.007	-.611	-1.336	.274	-.611	.512
Water	-.414	-.706	-1.788	.172	-.718	.685

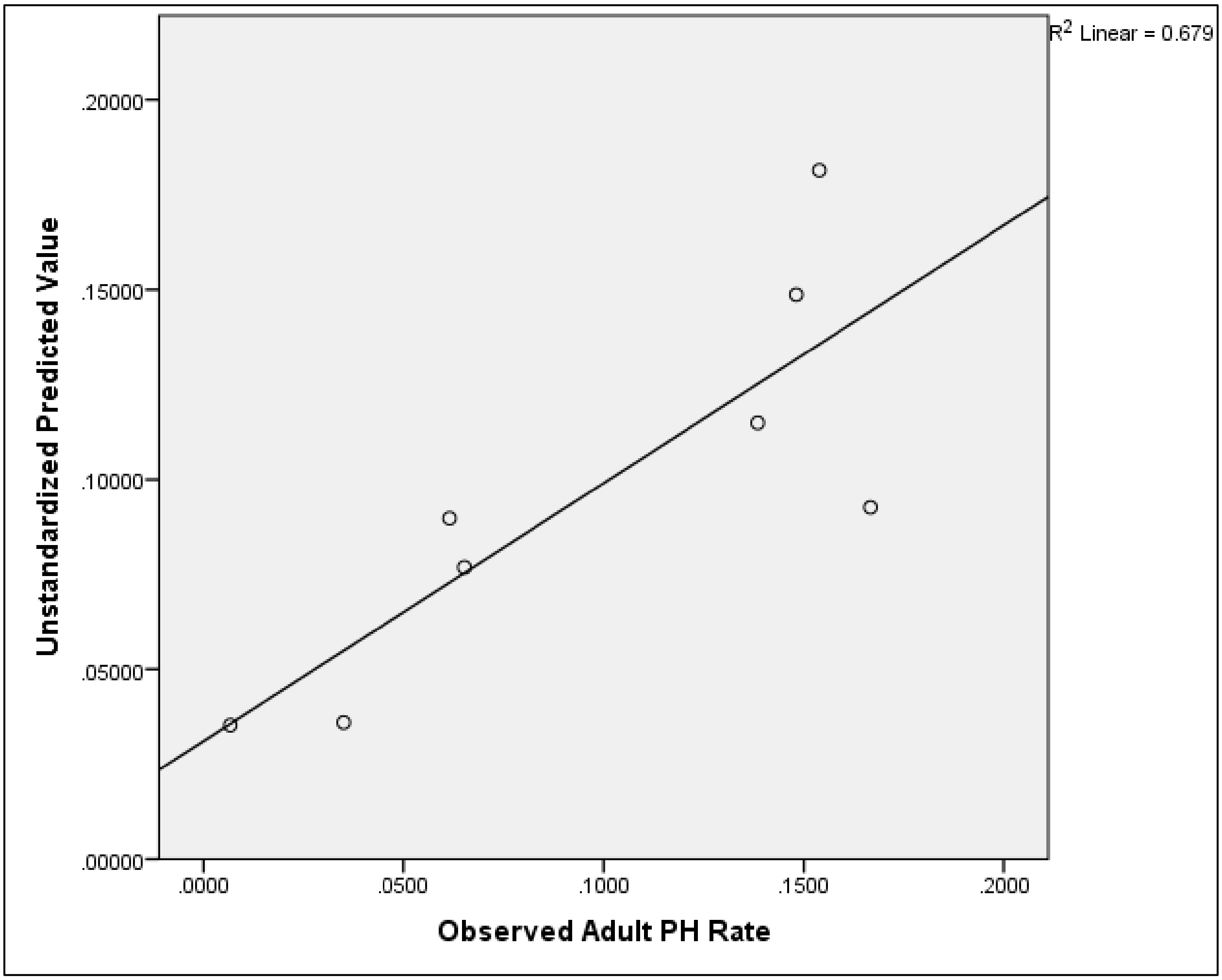

Figure 9. Plot of predicted adult porotic hyperostosis rate against the observed.

Table 11. Results of multiple linear regression for cribra orbitalia in adults.

N	**R^2**	**Standard Error of the Estimate**				
8	.727	.0355716				
	Unstandardized Coefficient	**Standardized Coefficient**	**t**	**p**	**Partial Correlations**	**Collinearity Tolerance**
Constant	-.216		-1.530	.223		
Soil Drainage	.018	.148	.347	.751	.197	.503
Temperature	-.002	-.263	-.545	.624	-.300	.393
Precipitation	.007	.851	2.017	.137	.759	.512
Water	.128	.303	.832	.467	.433	.685

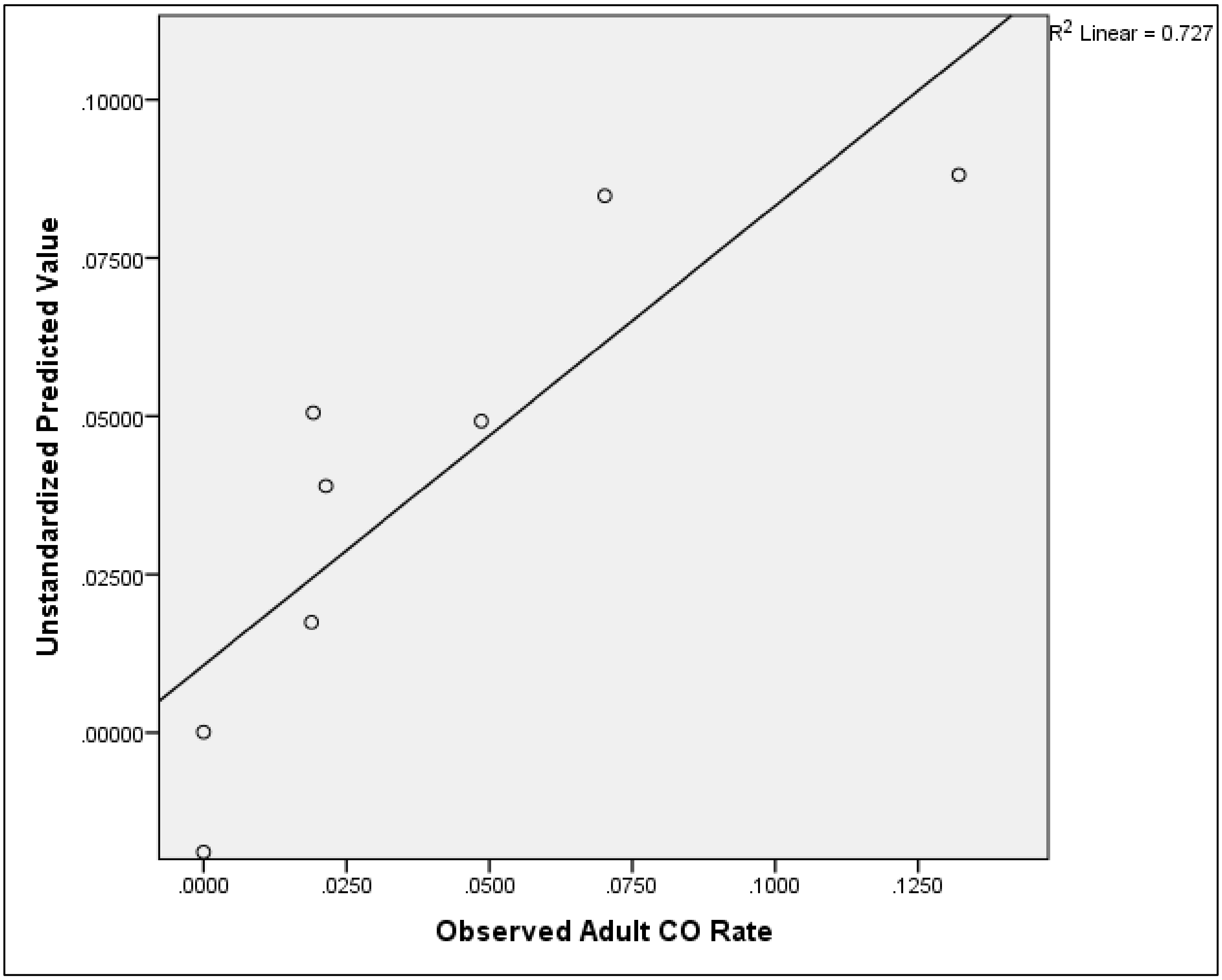

Figure 10. Plot of predicted adult cribra orbitalia rate against the observed.

5.4 Correlation with $\delta^{13}C$

Below are the results of correlation and regression tests using $\delta^{13}C$ to predict lesion rates for adults and children.

5.4.1 Adults and $\delta^{13}C$

As discussed earlier, Anderson was an outlier and was therefore removed from the sample. A square root transformation was applied. Using 18 degrees of freedom and an α of 0.05, the t distribution suggests that any studentized deleted residual value greater than 2.101 or less than -2.101 is an outlier (Anderson et al., 2014, p 660). Averbuch met this criterion (studentized deleted residual of 2.43) and was therefore removed from the sample. Simple linear regression gave insignificant ($R^2 = .000$, $p = 0.977$) results and Figure 11 demonstrates clearly the lack of a linear relationship. To investigate the possibility of a monotonic relationship, I ran a Spearman's correlation. The resulting correlation coefficient of -0.030 was not statistically significant ($p = 0.934$), suggesting that the null hypothesis of no monotonic correlation should be retained.

5.4.2 Children and $\delta^{13}C$

A square root transformation was applied. Using nine degrees of freedom and an α of 0.05, the t distribution suggests that any studentized deleted residual value greater than 2.26 or less than -2.26 is an outlier (Anderson et al., 2014, p 660). Norris Farms #36 met this criterion (studentized deleted residual of 2.76) and was therefore removed from the sample. Once Norris Farms #36 was removed from the sample and the residuals were reanalyzed, the studentized deleted residual for Anderson had a value of 3.20 and so this site was also removed as an outlier.

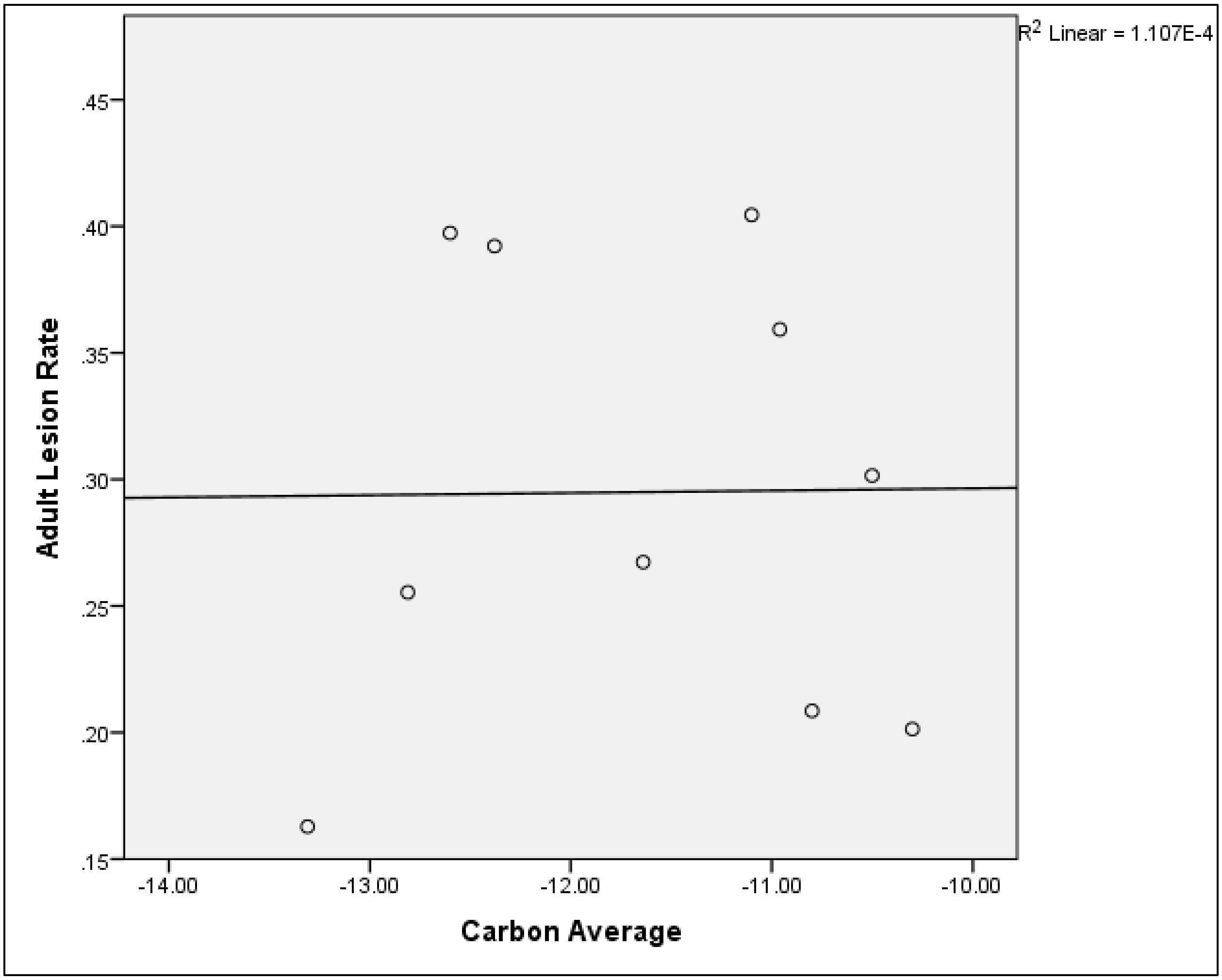

Figure 11. Relationship between adult lesion rate and $\delta^{13}C$ average.

Simple linear regression using $\delta^{13}C$ to predict lesion rates (n = 10) resulted in an R^2 value that says that 52.8% of the variability in lesion rates in children can be ‘explained’ by $\delta^{13}C$. These results were statistically significant with a p-value of 0.017, and Figure 12 demonstrates this relationship visually. The regression line equation was *predicted lesion rate* = 1.461 + (0.096 x $\delta^{13}C$). The results of Spearman’s correlation were also significant (p = 0.042), and the correlation coefficient of 0.829 suggests a strong positive relationship.

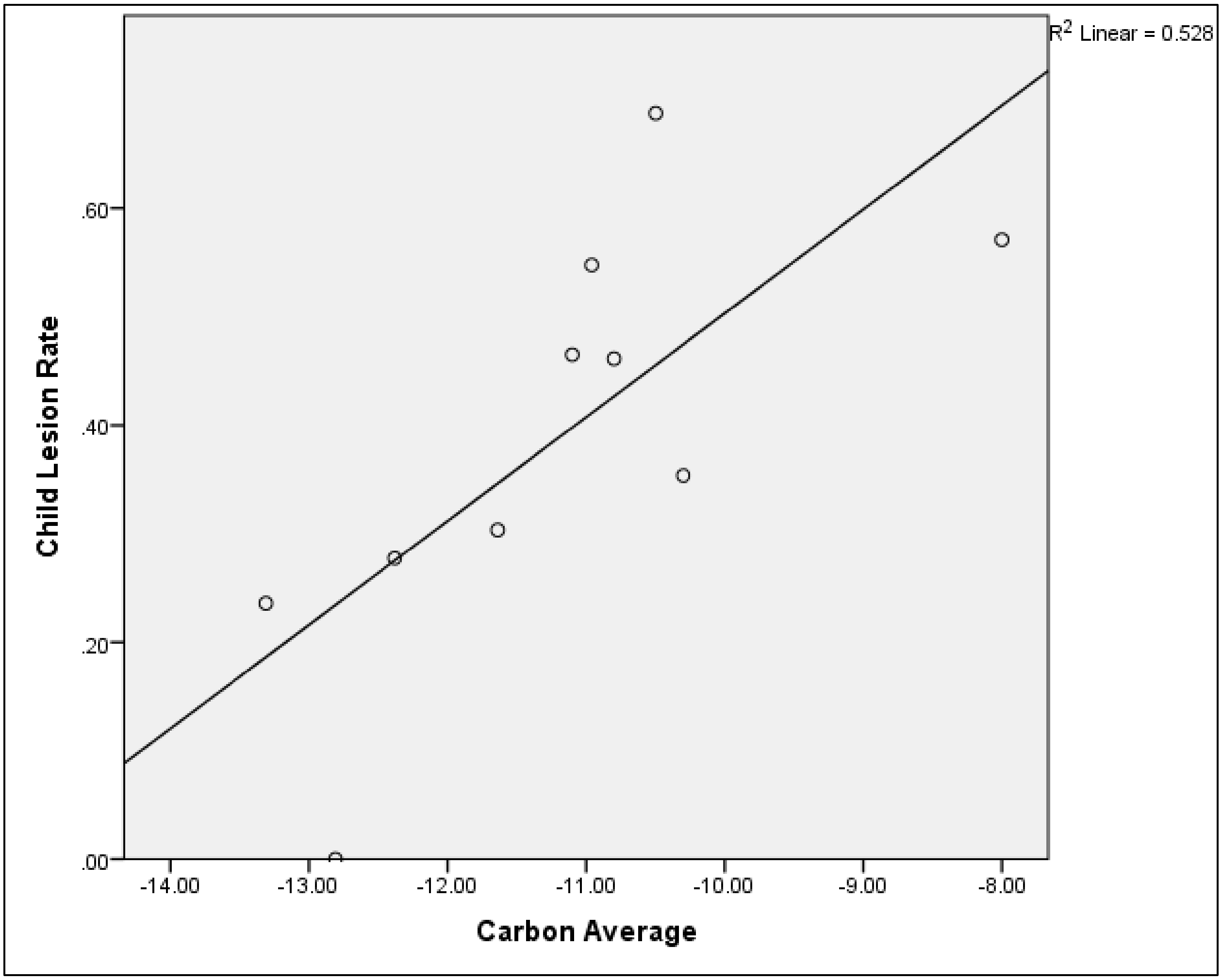

Figure 12. Relationship between child lesion rate and $\delta^{13}C$ average.

5.5 Comparison of prehistoric and historic data

Below are the results of linear regression using historic hookworm infection rates to predict lesion rates for adults and children, as well as the results of using the five environmental variables to predict historic hookworm infection rates.

5.5.1 Adult prehistoric lesion rate and historic infection rate

A square root transformation was applied to adult lesion rate, and no outliers were detected for the dependent variable before analysis or after analysis of studentized deleted residuals. Simple linear regression using historic hookworm infection rates to predict prehistoric adult lesion rates (*n* = 9) resulted in an R^2 value of 0.194 and a statistically insignificant p-value of 0.235. Figure 13 demonstrates the relationship visually. The regression line formula was *predicted lesion rate* = 0.131 + (0.653 x *historic infection rate*).

5.5.2 Child prehistoric lesion rate and historic infection rate

For children, a square root transformation was once again applied. Using three degrees of freedom and an α of 0.05, the t distribution suggests that any studentized deleted residual value greater than 3.181 or less than -3.181 is an outlier (Anderson et al., 2014, p 660). Cox met this criterion (studentized deleted residual of 4.24) and was therefore removed from the sample. Simple linear regression using historic hookworm infection rates to predict prehistoric lesion rates (*n* = 5) resulted in an R^2 value of 0.944 and a statistically significant p-value of 0.006. The relationship can be seen visually in Figure 14. The regression line formula was *predicted lesion rate* = -0.037 + (1.790 x *historic infection rate*).

5.5.3 Historic infection rate and environmental predictors

The results of multiple linear regression to predict historic rate of infection based on environmental variables can be seen in Table 12. Using three degrees of freedom and an α of 0.05, the t distribution suggests that any studentized deleted residual value greater than 3.181 or less than -3.181 is an outlier (Anderson et al., 2014, p 660). Boytt's Field met this criterion (studentized deleted residual of 3.65) and was therefore removed from the sample. When all five environmental variables were included, the collinearity tolerance value for temperature was too low (0.071) and this variable was therefore removed from the model. The four variable model

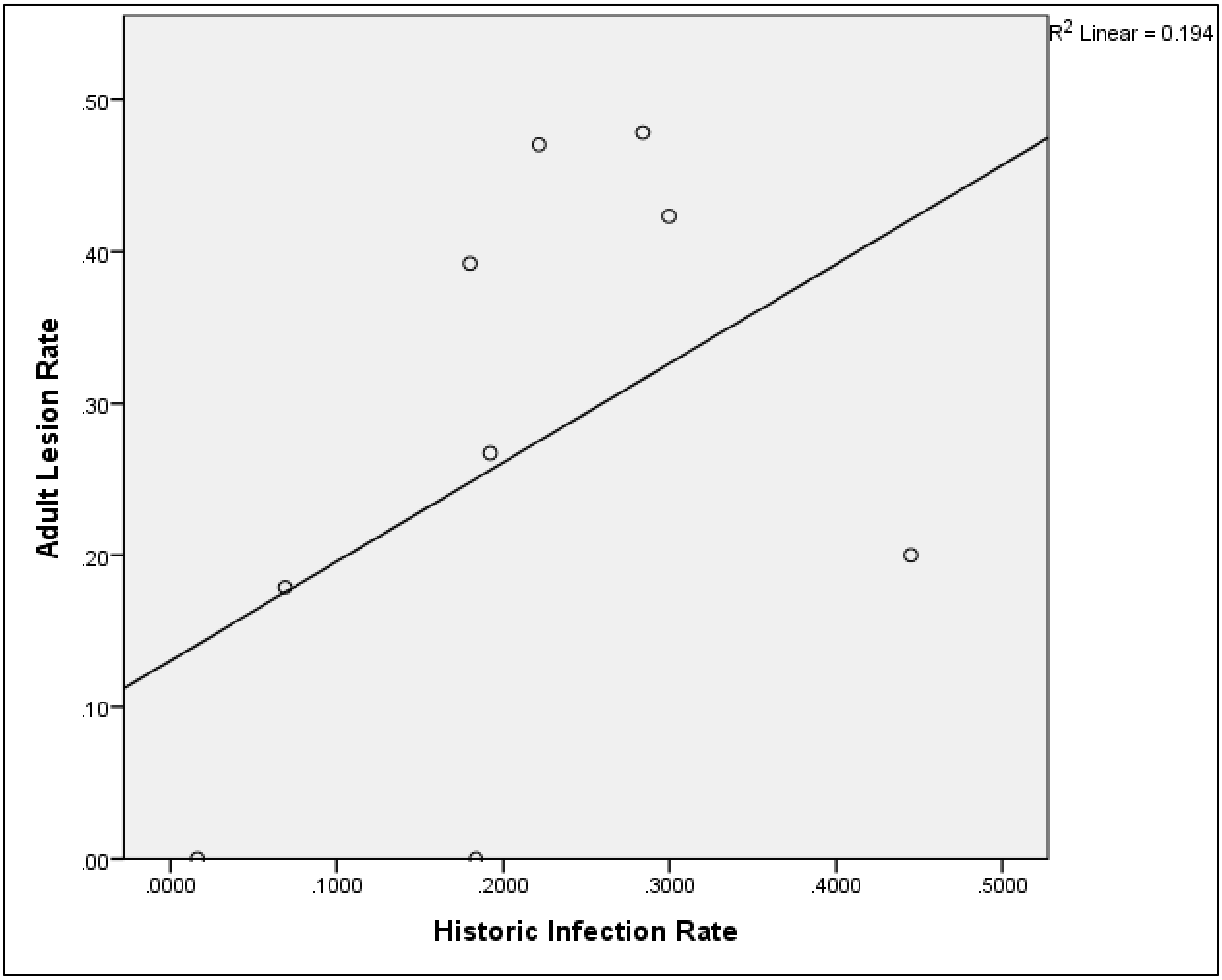

Figure 13. Relationship between adult lesion rate and historic infection rate.

resulted in an R^2 value of 0.450, and a statistically insignificant ANOVA p-value of 0.575. The relationships with soil drainage, elevation and precipitation were positive, and the relationship with the surface area of water was negative. Precipitation, soil drainage and surface area of water were the most influential variables. The predicted and observed lesion rates are plotted against each other in Figure 15.

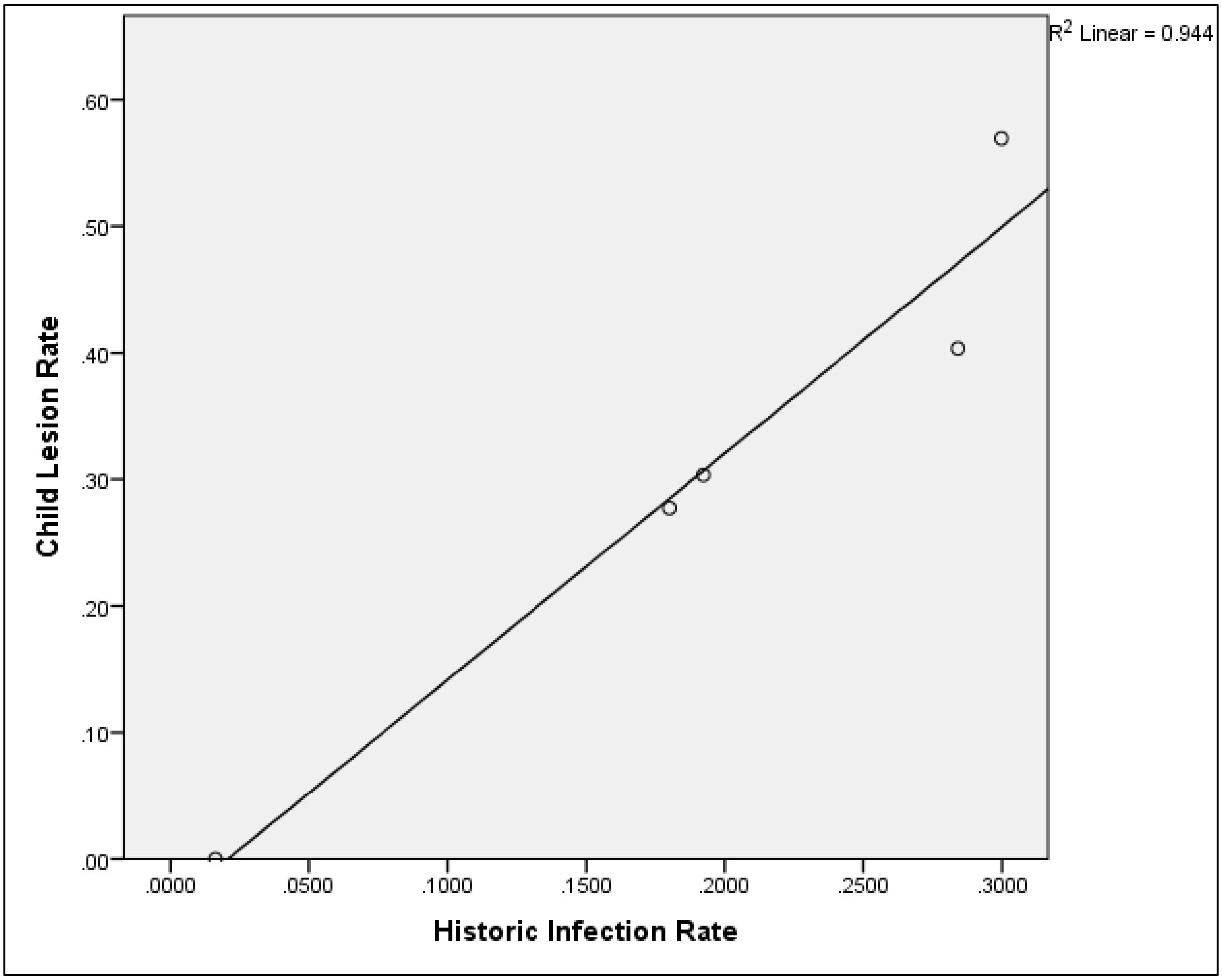

Figure 14. Relationship between child lesion rate and historic infection rate.

Table 12. Results of multiple linear regression for historic hookworm infection.

N	R^2	Standard Error of the Estimate	p
9	.450	.1453426	.575

	Unstandardized Coefficient	Standardized Coefficient	t	p	Partial Correlations	Collinearity Tolerance
Constant	-.418		-.880	.429		
Soil Drainage	.188	.606	.637	.559	.303	.152
Elevation	.000	.313	.355	.741	.175	.176
Precipitation	.009	.521	1.058	.350	.468	.567
Water	-1.294	-.440	-.727	.508	-.341	.376

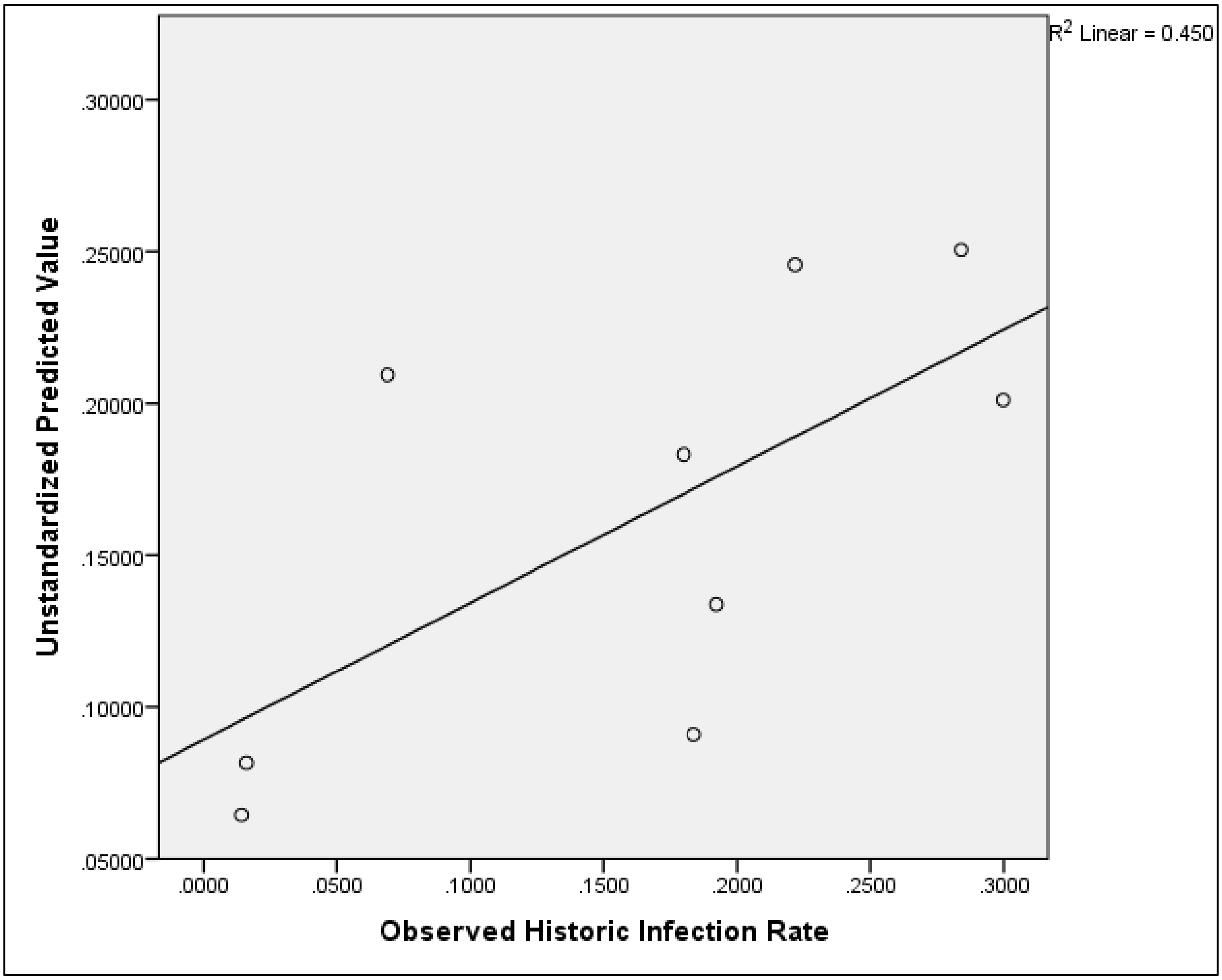

Figure 15. Plot of predicted historic infection rate against the observed.

5.6 Summary

The results of multiple linear regression was statistically significant for both adults and children, resulting in p-values of 0.040 and 0.029, respectively. The R^2 value was 0.532 for adults and 0.637 for children. The results of multiple linear regression looking at lesions separately for both age groups showed higher R^2 values than when lesions were combined, although small sample sizes precluded consideration of statistical significance. The direction of the relationship between the dependent and independent variables varied, with the exception of elevation and $\delta^{13}C$ which were always positive. Soil drainage, surface area of water and precipitation were most often the primary contributors to the model. Collagen carbon isostope values were able to successfully predict lesion rates using simple linear regression for children ($p = 0.017$) but not for adults ($p = 0.977$). Similarly, historic hookworm infection rates were successful in predicting prehistoric lesion rates using simple linear regression for children ($R^2 = 0.944$, $p = 0.006$) but not for adults ($R^2 = 0.194$, $p = 0.235$). The environmental variables were not able to successfully predict historic hookworm infection rates ($R^2 = 0.450$, $p = 0.575$).

6. DISCUSSION

Here I will discuss the possible meanings of the patterns described in Chapter 5, and compare these findings to the hypotheses laid out in Chapter 4. As previously mentioned, the analyses in which porotic hyperostosis and cribra orbitalia were separated for each age group were intended to be exploratory, and there was no significance testing given the small sample sizes. Table 13 summarizes the relationships between environmental variables and lesion rate for the various subgroups analyzed here, and compares these relationships to those expected for each parasite. It is not possible in all instances to discern which specific parasites are associated with observed correlations – for example, *G. lamblia* is in this analysis only predicted to have a relationship with precipitation, but precipitation is predicted to have the same relationship with *A. lumbricoides*, *T. trichiura*, and *N. americanus*. The two variables which may allow for particular parasites to be teased apart are (1) soil drainage, which has only been associated with *N. americanus*, and (2) temperature, for which the relationship is expected to be positive for the three soil transmitted helminths and negative for the three "water parasites". Following the interpretation of results, I will outline some of the limitations of this study and possibilities for future research.

6.1 General patterns observed

One of the most striking overall patterns here are the differences between adults and children. This was seen first in the results of chi-squared analysis, where lesion presence and age were related at a majority of sites, and for most of these the standardized residual for children with lesions was positive. This suggests that children usually have higher lesion rates than adults. With regard to the regression analyses, children met relationship expectations with the

Table 13. Summary of relationship directions in multiple linear regression.

Parasite	Ecological Variable	Expected Direction	Results					
			Adult	Child	Adult PH	Adult CO	Child PH	Child CO
Necator americanus	temperature	+	–	+	+	–	**–**	–
	precipitation	+	–	+	–	**+**	**+**	**+**
	soil drainage	+	**+***	**+**	**+**	+	–	+
	elevation	–	+	+	n/a	n/a	n/a	n/a
Trichuris trichiura	temperature	+	–	+	+	–	**–**	–
	precipitation	+	–	+	–	**+**	**+**	**+**
Ascaris lumbricoides	temperature	+	–	+	+	–	**–**	–
	precipitation	+	–	+	–	**+**	**+**	**+**
Echinostoma spp.	temperature	–	–	+	+	–	**–**	–
	water	+	–	**+***	**–**	+	**+**	**+**
Diphyllobothrium spp.	temperature	–	–	+	+	–	**–**	–
	water	+	–	**+***	**–**	+	**+**	**+**
Giardia lamblia	temperature	–	–	+	+	–	**–**	–
	precipitation	+	–	+	–	**+**	**+**	**+**

*significant at 0.05 level
shaded boxes are variables that meet expectations
bolded signs (+ or –) are instances where the variable was a top contributor to the model

environmental variables more often than adults, both when lesions were separated and when they were combined. Finally, child lesion rates correlated significantly with both $\delta^{13}C$ and historic infection rates while adult lesion rates did not.

The most likely explanation for this is differential healing in adults – that is, the healing of some lesions in some adults obscures the true rates of anemia that these individuals experienced during childhood. In contrast to children, adults did not correlate statistically significantly with either $\delta^{13}C$ or historic infection rate. Further, in the exploratory analyses where the two lesions were separated for adults, the lesions were very different from each other in terms of the strength and direction of relationships with the environmental variables. The only instance in which adult lesion rate showed statistically significant correlation was in the largest sample when the two lesions were combined. Given the differences in relationship patterning when the two lesions were separated, it is possible that the true form and strength of the relationship is obscured by combining the two lesions which may be, in fact, heterogeneous subgroups.

Another relatively consistent pattern here was the implication of *Necator americanus* as a potential cause of anemia. This was seen in the importance of soil drainage as a variable in most models, and that the direction of the relationship was usually in line with expectations. This is not a surprising finding given the results of the study by Anderson and Allen (2011) in an overlapping geographic area.

Also somewhat consistently implicated for children were the “water parasites,” *Echinostoma* spp., *Diphyllobothrium* spp. and *Giardia lamblia*. In both the sample where lesions were combined and the samples where the lesions were separated for children, there was general agreement with expectations regarding these parasites. Variables associated with these parasites were also usually important contributors to the models. It is possible that this pattern would also appear in the adult samples if healing was not obscuring the relationship; in fact, for cribra orbitalia in adults, the directions of relationships are indeed in line with expectations for the water parasites. It is also possible that children are more likely than adults to develop anemia from these kinds of parasites because they don’t have built up stores of iron and B_{12} in their bodies.

These observations are all suggestive, but there is also a great deal of inconsistency in the patterning with regard to the strength and the direction of relationships with variables, and the changes seen when the two lesions are separated. I would expect more consistency in subsamples if the correlations seen here really are due to parasite infection. A possible alternative explanation is that the relationship with the environmental variables is more direct; perhaps rather than acting as a proxy for parasite infection, these variables represent varying environmental conditions under which bone degenerates and creates pseudopathological "lesions" that look like cribra orbitalia or porotic hyperostosis, but are not actually the result of disease. Some of these individuals may have been misdiagnosed and the foramina on their crania may not in fact be the result of anemia.

This broaches another potential alternative explanation, which is that these lesions are due to disease but not always anemia. Osteomyelitis, rickets and scurvy have all been shown to cause lesions that can be similar in appearance (Schultz, 2003). If some lesions are due to these other diseases and not anemia, they could be obscuring the relationships with the environmental variables. Smaller samples are most likely to be affected by this, and this is the third possible explanation for the inconsistencies seen here. Small samples are more likely to be affected by outliers, and the addition or removal of a single case can alter a model significantly (Norušis, 2012).

6.2 Comparison of results to hypotheses

Above I discussed the general patterns observed and some possible explanations for them. Here I will discuss the results of analysis with regard to the initial six hypotheses formulated in Chapter 2.

Hypothesis 1: There will be significant correlation between prehistoric lesion rates and environmental variables

Support for this hypothesis is seen in the two largest samples, where both adult and child lesion rates demonstrated statistically significant correlation ($p = 0.040$ and 0.029, respectively) with the environmental variables. *Necator americanus* and the three "water parasites" were most consistently implicated, particularly for children. As previously discussed, the linear relationship seen with adult lesion rates may be an illusion resulting from combining porotic hyperostosis and cribra orbitalia in this sample, and a future study with larger sample sizes could potentially shed more light on this. Overall though, this hypothesis is supported.

Hypothesis 2: Children will show stronger correlation than adults

This hypothesis is also supported by the results of this analysis. Children had a higher R^2 value than adults (0.637 vs. 0.532), and they met expectations more consistently than adults in terms of the predicted direction of relationships with environmental variables. Further, child lesion rates were statistically significantly correlated with $\delta^{13}C$ and historic hookworm infection rates, while adult lesion rates were not. These findings are most likely the result of healing in some adults, resulting in lesion rates that do not accurately reflect the anemia these individuals experienced during childhood. However, when the lesions were separated, child lesion rates had the expected higher R^2 value for porotic hyperostosis, but adults had a higher R^2 value for cribra orbitalia. There is no clinical explanation for this, and it is possible that larger sample sizes would yield clearer patterning.

Hypothesis 3: Women will show stronger correlation than men

This hypothesis was not fully considered here after the result of chi-squared analysis demonstrated that sex and lesion presence were not related. However, it is worth noting that two of the four sites that were used to make this determination, Anderson and Averbuch, both ended up being eliminated as outliers from several analyses. Though the results here suggest that there is no significant difference between male and female lesion rates, it is possible that larger and/or different samples would yield different results. Though this hypothesis was not supported by this analysis, it bears further inquiry.

Hypothesis 4: Cribra orbitalia and porotic hyperostosis will correlate better separately than when grouped together

Though this hypothesis could not be explored rigorously with statistics due to small sample sizes, the results of exploratory multiple linear regression are suggestive. The R^2 values increased for both adults and children when lesions were separated – adults increased from 0.532 when combined to 0.679 for porotic hyperostosis and 0.727 for cribra orbitalia, while children increased from 0.532 when combined to 0.887 for porotic hyperostosis and 0.661 for cribra orbitalia. This suggests that these lesions may indeed have separate etiologies.

Hypothesis 5: $\delta^{13}C$ values will be able to successfully predict prehistoric lesion rates

The results of this analysis found this hypothesis to be true for child lesion rates (R^2 = 0.528, p = 0.017) but not for adult lesion rates (R^2 = .000, p = 0.977). As previously discussed, healing in adults is one possible explanation for this pattern. Another possible explanation is that the linear relationship seen for children is a fluke specific to this small sample (*n* = 10) and that in reality, neither adult nor child lesion rates have a linear relationship with $\delta^{13}C$. Preferential protein routing to bone collagen perhaps ‘drowns out’ the evidence for maize consumption at some of these sites, making the relationship monotonic but non-linear. The results of Spearman’s correlation were significant for children (p = 0.042) and not for adults (p = 0.934), and if this were true in another sample where linear regression was not successful, then the second explanation (preferential protein routing) would be supported.

Hypothesis 6: Historic rates of infection and prehistoric lesion rates will be similar

As with $\delta^{13}C$, the result of this analysis found this hypothesis to be true for child lesion rates ($R^2 = 0.944$, $p = 0.006$) but not for adult lesion rates ($R^2 = .194$, $p = 0.235$). This is likely a result of healing in adults, though the result for children could be specific to the small sample available here ($n = 5$). However, multiple linear regression using the five environmental variables to predict historic infection rates did not demonstrate a statistically significant linear relationship ($R^2 = 0.450$, $p = 0.575$). However, given the findings of Anderson and Allen (2011) in the same geographic area using the same historic dataset, it was surprising to see that soil drainage was not a more important variable in the model ($p = 0.559$). I suspect that the multiple linear regression model failed not because there is no relationship, but rather because the data are incompatible: the historic hookworm infection rates are at the county level, while the environmental values are specific to a 15 km radius around an archaeological site within that county. Overall, the results here provide another line of support for *N. americanus* as a cause of anemia from parasite infection in the past.

6.3 Summary

The results of analysis for each hypothesis are summarized in Table 14. Statistically significant correlation was found for both adults and children between prehistoric lesion rates and ecological variables. Smaller samples, such as when the lesions were examined separately, often yielded statistically insignificant results. The strength and direction of relationships showed a great deal of variation between groups, though *N. americanus* and the three “water parasites” (*Echinostoma* spp., *Diphyllobothrium* spp. and *G. lamblia*) were most consistently implicated. The observed correlations may be due to parasite infection, though I would expect that the

Table 14. Summary of general hypotheses, results and conclusions.

Hypothesis	Results	Conclusion
There will be significant correlation between prehistoric lesion rates and environmental variables	(1) Correlation found between ecological variables and lesion rates in both children and adults (2) Relationships inconsistent across subgroups, so other explanations possible	partially supported
Children will show stronger correlation than adults	(1) Higher R^2 value for children than for adults (2) Relationships more often consistent with expectations for children than adults (3) Linear relationship found between $\delta^{13}C$ and historic infection rate for children but not adults	supported
Women will show stronger correlation than men	(1) Chi-squared results demonstrated insignificant differences in lesion rates	not supported
Cribra orbitalia and porotic hyperostosis will correlate better separately than when grouped together	(1) Higher R^2 values for each lesion separately than for when combined (2) Results statistically insignificant	partially supported
$\delta^{13}C$ values will be able to successfully predict prehistoric lesion rates	(1) Linear regression significant for children but not adults	partially supported
Historic rates of infection and prehistoric lesion rates will be similar	(1) Historic rates could predict child lesion rate but not adult lesion rate in simple linear regression (2) Environmental variables could not predict historic hookworm infection rates, though likely the result of incompatible data	partially supported

strength and direction of relationships with ecological variables would remain more consistent for each subgroup. However, the inconsistencies between children and adults could be the result of differential healing in adults, and the inconsistencies when lesions were separated could be the result of small sample sizes. Small samples are easily affected by outliers and may not accurately reflect the true relationships found in the populations they represent.

6.4 Limitations

There are of course a number of complicating factors, and any conclusions drawn should be viewed as preliminary. Sample size has already been mentioned as one of these issues. Further, in an ideal study, scale and comparability of samples would be controlled for. Scale matters because even when laboratory studies show that an ecological variable influences the life-cycle of a parasite, it may be in such a way that a pattern would not be detectable using relatively coarse-grained satellite data. For example, a satellite will assign an average temperature value to a 30 by 30 meter square, but in reality there is a range of temperatures within that area. A parasite that thrives between 28 and 30°C will not be predicted ‘present’ within a square with the value 33°C (an average), even if that square in reality contains 15 by 30 meters of livable (29°C) and 15 by 30 meters non-livable (37°C) temperatures.

It is also potentially limiting to use modern climate data to predict prehistoric outcomes, as it forces reliance on an assumption that may not be true – in this case, that the differences in variables like temperature between location A and location B are the same now as they were in the past. This assumption is probably safest for the variables elevation and soil drainage which are minimally impacted by industrialization, and riskiest for variables like temperature given the climate change that is occurring worldwide today.

The uniformity of samples is also not ideal because data on porotic hyperostosis, cribra orbitalia, stable isotope ratios, and clinical hookworm infection were collected in different ways by different researchers. Interobserver error is well-known concern in bioarchaeology, and as previously discussed in Chapter 2, the issue was examined with regard to porotic hyperostosis and cribra orbitalia in a study by Jacobi and Danforth (2002). Stable isotope analysis is somewhat less subjective, though the lack of a standardized laboratory procedure for sample processing (c.f. differences in the physical state of bone, demineralization, solvent, alkalai, gelatinization and filtration used in Katzenberg and Lovell, 1999; Harbeck and Grupe, 2009; Caputo et al., 2012; Mays and Beavan, 2012) may lead to differences in the final ratios observed.

As previously mentioned, the use of porotic hyperostosis and cribra orbitalia as indicators of anemia is further complicated by the possibility that, along with post-mortem bone degenerative processes, they can sometimes result from diseases and illnesses other than anemia.

While visual examination remains the most popular method for diagnosis of porotic hyperostosis and cribra orbitalia, microscopic studies have shown that lesions that superficially resemble porotic hyperostosis and cribra orbitalia can result from many diseases, including osteomyelitis, anemia and rickets (Schultz, 2001).

A further caveat is that the individuals at each of these sites may not all have been of the same social status. This could have affected the kinds of activities an individual participated in, the foods they had access to, and the kind of care a person received when ill. All of these factors can have a significant influence on health. Ideally for a comparison like this, individuals both within and between sites would be of similar status. However, in reality this is a very difficult variable to control for. Identifying status from a burial is complicated and not always straightforward (Ucko, 1969), and even if individuals can be divided into status groups within a single site, 'high status' at one location may not imply the same privileges as 'high status' in another location. In this analysis I have included all individuals available at each site, but the possible influence of status on variation in lesion rates should be kept in mind.

Another potential issue is that the skeletal samples in question may not perfectly represent the populations from which they are derived. Wood et al. (1992) point out three problems for reconstruction of past health that had been previously overlooked, collectively known as the "osteological paradox". These three problems are demographic nonstationarity, selective mortality, and hidden heterogeneity in risks. It is this second problem that is most relevant here. Selective mortality is the idea that a skeletal sample only represents the people who died – that is, the only twenty year olds in a cemetery will be the twenty year olds who died at that age. Those who lived through age twenty might appear as a sixty year old instead. This means that younger skeletons with lesions may represent a larger proportion of young people in a sample than there would have been in that age group during life (that is, if their lesions were linked to their death). This can affect comparisons of subgroups like adults and children. In their original article, Wood et al. (1992) offer a few suggestions for how to deal with these problems, including the need to gain a better understanding of the biological processes behind skeletal lesions at the cellular level. This allows for better assessment of the etiology of skeletal lesions, and helps us understand how severe a particular illness has to be in order to remodel bone. More recently, Wright and Yoder (2003) discussed recent advances in dealing with the osteological paradox, including advances in techniques like biodistance analysis, DNA, stable isotopes and histology. Correct identification and assessment of skeletal lesions and their underlying causes are key for addressing issues like selective mortality, and greatly increase a researcher's ability to correctly interpret results.

Skeletal tissue-specific issues aside, many of these same problems exist in modern epidemiological studies. Surveys are often conducted by different researchers with different methods and aims, and variables like age and socioeconomic status are not always consistently recorded. Samples are often collected from places like schools, which demographically may not accurately represent the population as a whole (Brooker and Michael, 2000). Despite these limitations, modern epidemiological studies have revealed some consistent patterns of correlation between environment and parasite infection. If the accuracy of what is classed as a lesion resulting from anemia can be improved, this method shows promise for providing insight into parasite infection in the past.

6.5 Future research

The results of this analysis suggest that there is a relationship between environment and the occurrence of skeletal lesions. Whether this relationship is related to parasite infection, post-mortem diagenetic processes or something else altogether remains unclear. As more and improved data become available, these relationships can be examined in more detail. One obvious improvement would be larger sample sizes, particularly for subgroups. Initiatives like the Global History of Health Project (Steckel et al., 2002) are helping make comparable datasets like this widely available.

Another improvement would be more accurate diagnosis of porotic hyperostosis and cribra orbitalia. Macroscopic visual inspection is by far the most common method for analysis, despite the clear limitations of this approach. Future studies could benefit greatly from the use of thin-sectioning or μCT scanning in order to narrow down the possible etiologies of these lesions. Computed tomography in particular is promising since it is a non-destructive method. Microscopic analysis can allow for stronger conclusions and greater confidence in the results obtained. Though these techniques can be time consuming and more expensive, even using them on a selection of skulls can confirm the presence of hypothesized diseases such as anemia, rickets, and scurvy, as well as subperiosteal inflammation.

A further boon to future studies would be stable isotope ratios of not just bone collagen, but also that of bone apatite. Controlled diet studies have shown that bone apatite carbonate is better than collagen at representing whole diet (Lee-Thorp, 2008). This is because the carbonate in bone comes from bicarbonate that is dissolved in blood, which is derived from the whole diet. This would eliminate the problem of non-linearity in the relationship between maize consumption and collagen $\delta^{13}C$ values. Including this data in the model could shed light on the role of diet in causing acquired anemia.

A final approach that could be used to study this topic would be to create a predictive model and test prehistoric lesion rate data against this. It could be difficult to get a prehistoric sample size large enough to both create and test the model, but historic data like that collected by the RSC could potentially be used to create the model and then the prehistoric data could be used to test it. This kind of approach is becoming more and more common in epidemiological studies (Basáñez et al., 2004). This method could at least be used to establish the expected strength and direction of relationships with ecological variables in the specific geographic area of study, something that is not possible using modern data since parasite infections like hookworm are rare in the study area today thanks to modern medicine.

7. CONCLUSION

This analysis set out to address whether or not parasite infection was a cause of anemia in the prehistoric eastern United States. Rates of porotic hyperostosis and cribra orbitalia for children and adults were recorded from twenty-two sites in the eastern United States. Environmental data was collected from each of these sites using GIS, and these were intended to represent risk for parasite infection.

As expected, there was statistically significant correlation between ecological variables and skeletal lesion rates for both adults and children. However, the direction of relationships varied unexpectedly between subgroups, suggesting that the correlations may not be the result of parasite infection. A possible alternative explanation is that the lesions are a result of post-mortem degenerative processes. Overall, the mechanism underlying the observed relationships remains unclear.

Comparison of children and adults showed stronger correlation for children, as expected. However, when observing the lesions separately, this pattern was not consistent. Porotic hyperostosis and cribra orbitalia separately resulted in stronger correlations within each age group than when they were combined. Larger samples are required to consider statistical significance and investigate these issues further.

Finally, comparison of males and females through chi-squared tests demonstrated no statistically significant differences in their lesion rates. The hypothesis that parasite infection affected females more than males was not supported.

In sum, it is clear that ecological variables and skeletal lesions are correlated in this sample, but the causal mechanism underlying this correlation is still unclear. Larger sample sizes would allow for more robust statistical analyses of the trends observed here. Relationship strength and direction demonstrated a great deal of variation between groups, but it is possible that larger samples would be less affected by outliers and would show more consistent patterning.

Despite the ambiguity of the specific patterns observed here, this study contributes to the body of literature demonstrating that porotic hyperostosis and cribra orbitalia cannot automatically be attributed to iron-deficiency anemia resulting from dependence on maize. One must consider not just what people did (e.g. agriculture) but also consider the environmental and social context within which they acted. Bioarchaeological studies are uniquely positioned for looking at the long-term effects of the natural and social environments on human health, and no doubt future research in this area will help us gain a better understanding of the factors which heightened and lessened risk for parasitic infection in the past.

References

Ambrose SH, Butler BM, Hanson DB, Hunter-Anderson RL, Krueger HW. 1997. Stable isotopic analysis of human diet in the Marianas Archipelago, Western Pacific. Am J Phys Anthropol 104:343–361.

Ambrose SH, Norr L. 1993. Experimental evidence for the relationship of the carbon isotope ratios of whole diet and dietary protein to those of bone collagen and carbonate. In: Prehistoric human bone. Springer. p 1–37.

Anderson AL, Allen T. 2011. Mapping Historic Hookworm Disease Prevalence in the Southern US, Comparing Percent Prevalence with Percent Soil Drainage Type Using GIS. Infect Dis Res Treat.

Anderson D, Sweeney D, Williams T, Camm J, Cochran J. 2014. Statistics for Business & Economics, Revised. Cengage Learning.

Angel JL. 1966. Porotic hyperostosis, anemias, malarias, and marshes in the prehistoric eastern Mediterranean. Science 153:760–763.

Le Bailly M, Bouchet F. 2013. Diphyllobothrium in the past: review and new records. Int J Paleopathol 3:182–187.

Basáñez M-G, Marshall C, Carabin H, Gyorkos T, Joseph L. 2004. Bayesian statistics for parasitologists. Trends Parasitol 20:85–91.

Ben-David M, Flaherty EA. 2012. Stable isotopes in mammalian research: a beginner’s guide. J Mammal 93:312–328.

Berryman HE. 1981. The Averbuch skeletal series a study of biological and social stress at a late Mississippian period site from Middle Tennessee.

Bethony J, Brooker S, Albonico M, Geiger SM, Loukas A, Diemert D, Hotez PJ. 2006. Soil-transmitted helminth infections: ascariasis, trichuriasis, and hookworm. The Lancet 367:1521–1532.

Blakely RL. 1980. Sociocultural implications of pathology between the village area and Mound C skeletal remains from Etowah, Georgia. Skelet Biol Aborig Popul Southeast U S Tenn Anthropol Assoc Misc Pap:28–38.

Blakely RL. 1995. Social organization at Etowah: A reconsideration of paleodemographic and paleonutritional evidence. Southeast Archaeol 14:46–59.

Blitz JH. 2010. New perspectives in Mississippian archaeology. J Archaeol Res 18:1–39.

Blom DE, Buikstra JE, Keng L, Tomczak PD, Shoreman E, Stevens-Tuttle D. 2005. Anemia and childhood mortality: Latitudinal patterning along the coast of pre-Columbian Peru. Am J Phys Anthropol 127:152–169.

Bogitsh BJ, Carter CE, Oeltmann TN. 2005. Human Parasitology. 3rd ed. Amsterdam: Elsevier.

Boyd DC. 1986. A comparison of Mouse Creek–phase to Dallas and middle Cumberland culture skeletal remains. In: Levy JE, editor. Skeletal analysis in Southeastern archaeology. Vol. 24. North Carolina Archaeological Council Publication. North Carolina Archaeological Council and the Archaeology Branch Archaeology and Historic Preservation Section North Carolina Division of Archives and History. p 103–127.

Boyd Jr CC, Boyd DC. 1991. A multidimensional investigation of biocultural relationships among three late prehistoric societies in Tennessee. Am Antiq:75–88.

Britton E, Hales S, Venugopal K, Baker M. 2010. The impact of climate variability and change on cryptosporidiosis and giardiasis rates in New Zealand. J Water Health 8:561–571.

Broida M. 1984. An estimate of the percents of maize in the diets of two Kentucky Fort Ancient villages. Late Prehist Res Ky Ky Herit Counc Frankf:68–82.

Brooker S, Clements AC, Bundy DA. 2006. Global epidemiology, ecology and control of soil-transmitted helminth infections. Adv Parasitol 62:221–261.

Brooker S, Michael E. 2000. The potential of geographical information systems and remote sensing in the epidemiology and control of human helminth infections. Adv Parasitol 47:245–288.

Buikstra JE, Milner GR. 1991. Isotopic and archaeological interpretations of diet in the Central Mississippi Valley. J Archaeol Sci 18:319–329.

Buikstra JE, Ubelaker DH. 1994. Standards for data collection from human skeletal remains. Fayetteville, AR.

Burkhard MJ, Brown DE, McGrath JP, Meador VP, Mayle DA, Keaton MJ, Hoffman WP, Zimmermann JL, Abbott DL, Sun SC. 2001. Evaluation of the erythroid regenerative response in two different models of experimentally induced iron deficiency anemia. Vet Clin Pathol 30:76–85.

Bush H. 2012. Biostatistics: An Applied Introduction for the Public Health Practitioner. Clifton Park, New York: Delmar: Cengage Learning.

Caldwell JR, McCann C, Hulse FS. 1941. Irene Mound Site, Chatham County, Georgia. The University of Georgia Press.

Caputo I, Lepretti M, Scarabino C, Esposito C, Proto A. 2012. An acetic acid-based extraction method to obtain high quality collagen from archeological bone remains. Anal Biochem 421:92–96.

Carlson DS, Armelagos GJ, Van Gerven DP. 1974. Factors influencing the etiology of cribra orbitalia in prehistoric Nubia. J Hum Evol 3:405–410.

Carrilho Galvao F, Da Costa Gileno M, Olivi Malta J, Vicentini V, Anibal F de F. 2011. Anemia in patients with intestinal parasitic infection. Rev Ibero-Latinoam Parasitol 70:206–211.

Carvalho-Costa FA, Gonçalves AQ, Lassance SL, Neto S, Salmazo CAA, Bóia MN. 2007. Giardia lamblia and other intestinal parasitic infections and their relationships with nutritional status in children in Brazilian Amazon. Rev Inst Med Trop São Paulo 49:147–153.

Cassidy CM. 1972. A comparison of nutrition and health in pre-agricultural and agricultural Amerindian skeletal populations.

Cassidy CM. 1984. Skeletal Evidence for Prehistoric Subsistence Adaptation in the Central Ohio River Valley. In: Paleopathology at the Origins of Agriculture. New York: Academic Press. p 307–345.

CDC. 2014. Parasites. Available from: http://www.cdc.gov/parasites/

Chase JW. 1988. A comparison of signs of nutritional stress in prehistoric populations of the Potomac Piedmont and coastal plain.

Cifuentes E, Suarez L, Espinosa M, Juarez-Figueroa L, Martinez-Palomo A. 2004. Risk of Giardia intestinalis infection in children from an artificially recharged groundwater area in Mexico City. Am J Trop Med Hyg 71:65–70.

Clay RB. 1997. The Mississippian succession on the lower Ohio. Southeast Archaeol 16:16–32.

Cook RA, Schurr MR. 2009. Eating between the lines: Mississippian migration and stable carbon isotope variation in Fort Ancient populations. Am Anthropol 111:344–359.

Cox FEG. 1993. Modern Parasitology: A Textbook of Parasitology. 2nd ed. Oxford: Blackwell.

DeNiro MJ, Epstein S. 1981. Influence of diet on the distribution of nitrogen isotopes in animals. Geochim Cosmochim Acta 45:341–351.

deRegnier DP, Cole L, Schupp DG, Erlandsen SL. 1989. Viability of Giardia cysts suspended in lake, river, and tap water. Appl Environ Microbiol 55:1223–1229.

Drake A, Oxenham M. 2013. Disease, climate and the peopling of the Americas. Hist Biol Int J Paleobiology 25:565–597.

Eisenberg LE. 1991. Mississippian Cultural Terminations in Middle Tennessee: What the Bioarchaeological Evidence Can Tell Us. In: Powell ML, Bridges PS, Mires AMW, editors. What Mean These Bones?: Studies in Southeastern Bioarchaeology. Tuscaloosa: University of Alabama Press. p 70–88.

El-Najjar MY, Ryan DJ, Turner CG, Lozoff B. 1976. The etiology of porotic hyperostosis among the prehistoric and historic Anasazi Indians of southwestern United States. Am J Phys Anthropol 44:477–487.

Emerson TE, Hargrave E. 2000. Strangers in paradise? Recognizing ethnic mortuary diversity on the fringes of Cahokia. Southeast Archaeol 19:1–23.

Erickson TR, Stefan HG. 2000. Linear air/water temperature correlations for streams during open water periods. J Hydrol Eng 5:317–321.

Evans N. 1985. The influence of environmental temperature upon transmission of the cercariae of Echinostoma liei (Digenea: Echinostomatidae). Parasitology 90:269–275.

Farid Z, Patwardhan VN, Darby WJ. 1969. Parasitism and anemia. Am J Clin Nutr 22:498–503.

Farrow DC. 1986. A study of Monongahela subsistence patterns based on mass spectrometric analysis. Midcont J Archaeol 11:153–179.

Foley JA, DeFries R, Asner GP, Barford C, Bonan G, Carpenter SR, Chapin FS, Coe MT, Daily GC, Gibbs HK, Helkowski JH, Holloway T, Howard EA, Kucharik CJ, Monfreda C, Patz JA, Prentice IC, Ramankutty N, Snyder PK. 2005. Global Consequences of Land Use. Science 309:570–574.

Galea JC. 2013. Analysing the microarchitecture of cribra orbitalia via micro-computed tomography in post-medieval remains from the Bristol Royal Infirmary.

Gold DL. 2004. The bioarchaeology of Virginia burial mounds. Tuscaloosa: University of Alabama Press.

Gonçalves MLC, Araújo A, Ferreira LF. 2003. Human intestinal parasites in the past: new findings and a review. Mem Inst Oswaldo Cruz 98:103–118.

Goodman AH, Lallo J, Armelagos GJ, Rose JC. 1984. Health Changes at Dickson Mounds, Illinois. In: Cohen MN, Armelagos GJ, editors. Paleopathology at the Origins of Agriculture. New York: Academic Press. p 271–305.

Gowland RL, Western AG. 2012. Morbidity in the marshes: Using spatial epidemiology to investigate skeletal evidence for malaria in Anglo-Saxon England (AD 410–1050). Am J Phys Anthropol.

Grauer AL. 2008. Macroscopic Analysis and Data Collection in Palaeopathology. In: Pinhasi R, Mays S, editors. Advances in human palaeopathology. John Wiley & Sons, Ltd. p 57–76.

Gray CD, Kinnear PR. 2012. IBM SPSS statistics 19 made simple. Psychology Press.

Griffin JB. 1966. The Fort Ancient Aspect: Its Cultural and Chronological Position in Mississippi Valley Archaeology. Ann Arbor: University of Michigan, Museum of Anthropology.

Griffin JB. 1992. Fort Ancient has no class: The absence of an elite group in Mississippian societies in the central Ohio Valley. Archeol Pap Am Anthropol Assoc 3:53–59.

Hanson LH. 1975. The Buffalo Site - A Late 17th Century Indian Village Site (46Pu31) in Putnam County, West Virginia. Morgantown: West Virginia Geological Survey.

Harbeck M, Grupe G. 2009. Experimental chemical degradation compared to natural diagenetic alteration of collagen: implications for collagen quality indicators for stable isotope analysis. Archaeol Anthropol Sci 1:43–57.

Harle MS. 2010. Biological affinities and the construction of cultural identity for the proposed Coosa Chiefdom.

Harris R, Jarvis C. 2014. Statistics for geography and environmental science. Routledge.

Hartson RB, Orlofske SA, Melin VE, Dillon Jr RT, Johnson PT. 2011. Land use and wetland spatial position jointly determine amphibian parasite communities. EcoHealth 8:485–500.

Hedges RE, Clement JG, Thomas CDL, O'Connell TC. 2007. Collagen turnover in the adult femoral mid-shaft: Modeled from anthropogenic radiocarbon tracer measurements. Am J Phys Anthropol 133:808–816.

Hedman K, Hargrave EA, Ambrose SH. 2002. Late Mississippian diet in the American Bottom: stable isotope analyses of bone collagen and apatite. MCJA Midcont J Archaeol 27:237–271.

Helms LJ. 2012. Health and Disease at Ledford Island: A Study of Late-Mississippian Human Remains.

Hogg RV, Tanis EA. 2005. Probability and Statistical Inference. 7th edition. Upper Saddle River, NJ: Prentice Hall.

Jacobi KP, Danforth ME. 2002. Analysis of interobserver scoring patterns in porotic hyperostosis and cribra orbitalia. Int J Osteoarchaeol 12:248–258.

Jiménez FA, Gardner SL, Araújo A, Fugassa M, Brooks RH, Racz E, Reinhard KJ. 2012. Zoonotic and Human Parasites of Inhabitants of Cueva de Los Muertos Chiquitos, Rio Zape Valley, Durango, Mexico. J Parasitol 98:304–309.

Johnson PT, McKenzie VJ. 2009. Effects of environmental change on helminth infections in amphibians: exploring the emergence of Ribeiroia and Echinostoma infections in North America. In: Fried B, Toledo R, editors. The biology of echinostomes: from the molecule to the community. New York: Springer. p 249–280.

Kassabaum MC. 2011. Looking Beyond the Obvious: Identifying Patterns in Coles Creek Mortuary Data. Southeast Archaeol 30:215–225.

Katzenberg MA, Lovell NC. 1999. Stable isotope variation in pathological bone. Int J Osteoarchaeol 9:316–324.

Kent S. 1986. The influence of sedentism and aggregation on porotic hyperostosis and anaemia: a case study. Man:605–636.

Lallo J. 1979. Disease and Mortality at the Anderson Site. Ohio J Sci 79:256–261.

Lallo JW, Armelagos GJ, Mensforth RP. 1977. The role of diet, disease, and physiology in the origin of porotic hyperostosis. Hum Biol 49:471–483.

Lallo JW, Blank JE. 1977. Ancient Disease in Ohio: The Eiden Population. Ohio 77:55–62.

Lane L. 1993. A Bioarchaeological Analysis of the Human Skeletal Population from Tinsley Hill (15Ly18a&b).

Larsen CS ed. 2001. Bioarchaeology of Spanish Florida: The impact of colonialism. Gainesville, Florida: University Press of Florida.

Larsen CS, Crosby AW, Griffin MC, Hutchinson DL, Ruff CB, Russell KF, Schoeninger MJ, Sering LE, Simpson SW, Takács JL, Teaford MF. 2002. A biohistory of health and behavior in the Georgia Bight. In: The backbone of history: health and nutrition in the Western Hemisphere. New York: Cambridge University Press. p 406–440.

Larsen CS, Hutchinson DL, Schoeninger MJ, Norr L. 2001. Food and Stable Isotopes in La Florida: diet and nutrition before and after contact. In: Larsen CS, editor. Bioarchaeology of Spanish Florida: The Impact of Colonialism. Gainesville, Florida: University Press of Florida. p 52–81.

Lee-Thorp JA. 2008. On Isotopes and Old Bones. Archaeometry 50:925–950.

Leles D, Araújo A, Ferreira LF, Vicente ACP, Iñiguez AM. 2008. Molecular paleoparasitological diagnosis of Ascaris sp. from coprolites: new scenery of ascariasis in pre-Colombian South America times. Mem Inst Oswaldo Cruz 103:106–108.

Leles D, Reinhard KJ, Fugassa M, Ferreira LF, Iñiguez AM, Araújo A. 2010. A parasitological paradox: Why is ascarid infection so rare in the prehistoric Americas? J Archaeol Sci 37:1510–1520.

Lovell NC. 1997. Anaemia in the ancient Indus Valley. Int J Osteoarchaeol 7:115–123.

Mays S, Beavan N. 2012. An investigation of diet in early Anglo-Saxon England using carbon and nitrogen stable isotope analysis of human bone collagen. J Archaeol Sci 39:867–874.

Mehta A, Hoffbrand V. 2013. Haematology at a Glance. 4th ed. Chichester: Wiley.

Metress JF. 1971. An Osteobiography of the Buffalo Site, Fulton County, West Virginia.

Mick CB. 2011. Status and Health at the Mississippian Period Averbuch Site.

Milner GR. 1983. The East St. Louis Stone Quarry Site Cemetery (11-S-468). Urbana and Chicago: University of Illinois Press.

Milner GR. 1991. Health and Cultural Change in the Late Prehistoric American Bottom, Illinois. In: Powell ML, Bridges PS, Mires AMW, editors. What Mean These Bones?: Studies in Southeastern Bioarchaeology. Tuscaloosa: University of Alabama Press. p 52–69.

Milner GR, Anderson E, Smith VG. 1991. Warfare in late prehistoric west-central Illinois. Am Antiq 56:581–603.

Milner GR, Humpf DA, Harpending HC. 1989. Pattern matching of age-at-death distributions in paleodemographic analysis. Am J Phys Anthropol 80:49–58.

Mohseni O, Stefan HG. 1999. Stream temperature/air temperature relationship: a physical interpretation. J Hydrol 218:128–141.

Montenegro A, Araujo A, Eby M, Ferreira LF, Hetherington R, Weaver AJ. 2006. Parasites, Paleoclimate, and the peopling of the Americas. Curr Anthropol 47:193–200.

Moore CB, Hrdlička A. 1909. Antiquities of the Ouachita Valley. Philadelphia: United States National Museum, P.C. Stockhausen.

Morgan JA, Dejong RJ, Adeoye GO, Ansa E, Barbosa CS, Bremond P, Cesari IM, Charbonnel N, Correa LR, Coulibaly G. 2005. Origin and diversification of the human parasite Schistosoma mansoni. Mol Ecol 14:3889–3902.

Norušis MJ. 2012. IBM SPSS Statistics 19 Guide to Data Analysis. New Jersey: Prentice Hall.

O'Connell TC, Kneale CJ, Tasevska N, Kuhnle GGC. 2012. The diet-body offset in human nitrogen isotopic values: A controlled dietary study. Am J Phys Anthropol 149:426–434.

Olivares JL, Fernández R, Fleta J, Ruiz MY, Clavel A. 2002. Vitamin B12 and folic acid in children with intestinal parasitic infection. J Am Coll Nutr 21:109–113.

Oski FA, Barness LA. 1967. Vitamin E deficiency: a previously unrecognized cause of hemolytic anemia in the premature infant. J Pediatr 70:211–220.

Oxenham MF, Cavill I. 2010. Porotic hyperostosis and cribra orbitalia: the erythropoietic response to iron-deficiency anaemia. Anthropol Sci 118:199–200.

Papathanasiou A. 2005. Health status of the Neolithic population of Alepotrypa Cave, Greece. Am J Phys Anthropol 126:377–390.

Parham KR, Scott GT. 1980. Porotic hyperostosis: A study of disease and culture at Toqua (40MR6), a late Mississippian site in eastern Tennessee. Skelet Biol Aborig Popul Southeast U S Tenn Anthropol Assoc Misc Pap:39–51.

Pechenkina EA, Delgado M. 2006. Dimensions of health and social structure in the early intermediate period cemetery at Villa El Salvador, Peru. Am J Phys Anthropol 131:218–235.

Peebles CS, Kus SM. 1977. Some archaeological correlates of ranked societies. Am Antiq 42:421–448.

Perry MA. 2005. Redefining childhood through bioarchaeology: Toward an archaeological and biological understanding of children in antiquity. Archeol Pap Am Anthropol Assoc 15:89–111.

Pilgrim J, Fang X, Stefan H. 1998. Stream temperature correlations with air temperatures in Minnesota: Implications for climate warming. Water Resour Bull 34:1109–1121.

Pollack D, Henderson AG, Begley CT. 2002. Fort Ancient/Mississippian interaction on the northeastern periphery. Southeast Archaeol:206–220.

Powell ML. 1991. Ranked Status and Health in the Mississippian Chiefdom at Moundville. In: Powell ML, Bridges PS, Mires AMW, editors. What Mean These Bones?: Studies in Southeastern Bioarchaeology. Tuscaloosa: University of Alabama Press. p 22–51.

Pullan R, Brooker S. 2008. The health impact of polyparasitism in humans: are we under-estimating the burden of parasitic diseases? Parasitology 135:783–794.

Raso G, Matthys B, N'goran EK, Tanner M, Vounatsou P, Utzinger J. 2005. Spatial risk prediction and mapping of Schistosoma mansoni infections among schoolchildren living in western Côte d'Ivoire. Parasitology 131:97–108.

Reinhard KJ. 1990. Archaeoparasitology in North America. Am J Phys Anthropol 82:145–163.

Reinhard KJ. 1992. Patterns of diet, parasitism, and anemia in prehistoric west North America. In: Stuart-Macadam P, Kent S, editors. Diet, Demography, and Disease: Changing Perspectives on Anemia. . p 219–258.

Reinhard KJ, Ferreira LF, Bouchet F, Sianto L, Dutra JMF, Iniguez A, Leles D, Le Bailly M, Fugassa M, Pucu E. 2013. Food, parasites, and epidemiological transitions: A broad perspective. Int J Paleopathol 3:150–157.

Reinhard KJ, Hevly RH, Anderson GA. 1987. Helminth remains from prehistoric Indian coprolites on the Colorado Plateau. J Parasitol 73:630–639.

Rezaeian M, Dunn G, St Leger S, Appleby L. 2007. Geographical epidemiology, spatial analysis and geographical information systems: a multidisciplinary glossary. J Epidemiol Community Health 61:98–102.

Rose JC, Burnett BA, Nassaney MS, Blaeuer MW. 1984. Paleopathology and the Origins of Maize Agriculture in the Lower Mississippi Valley and Caddoan Culture Areas. In: Cohen MN, Armelagos GJ, editors. Paleopathology at the Origins of Agriculture. New York: Academic Press. p 393–424.

Rose JC, Marks MK, Tieszen LL. 1991. Bioarchaeology and subsistence in the central and lower portions of the Mississippi Valley. In: Powell ML, Bridges PS, Mires AMW, editors. What Mean These Bones?: Studies in Southeastern Bioarchaeology. Tuscaloosa: University of Alabama Press. p 7–21.

Rothschild B. 2012. Extirpolation of the Mythology that Porotic Hyperostosis is Caused by Iron Deficiency Secondary to Dietary Shift to Maize. Adv Anthropol 2:157–160.

Roux AVD. 2004. The study of group-level factors in epidemiology: rethinking variables, study designs, and analytical approaches. Epidemiol Rev 26:104–111.

Salvadei L, Ricci F, Manzi G. 2001. Porotic hyperostosis as a marker of health and nutritional conditions during childhood: studies at the transition between Imperial Rome and the Early Middle Ages. Am J Hum Biol 13:709–717.

Schoeninger MJ, Schurr MR. 1998. Human subsistence at Moundville: the stable-isotope data. In: Knight V, Peebles C, Steponaitis V, editors. Archaeology of the Moundville Chiefdom: Chronology, Content, Contest. Tuscaloosa: University of Alabama Press.

Scholz T, Garcia HH, Kuchta R, Wicht B. 2009. Update on the human broad tapeworm (genus Diphyllobothrium), including clinical relevance. Clin Microbiol Rev 22:146–160.

Schroedl GF. 1998. Mississippian towns in the eastern Tennessee Valley. In: Lewis RB, Stout C, editors. Mississippian Towns and Sacred Spaces: Searching for an Architectural Grammar. University of Alabama Press. p 64–92.

Schultz M. 2001. Paleohistopathology of bone: a new approach to the study of ancient diseases. Am J Phys Anthropol 116:106–147.

Schultz M. 2003. Light microscopic analysis in skeletal paleopathology. In: Ortner DJ, editor. Identification of pathological conditions in human skeletal remains. Amsterdam: Academic Press. p 73–108.

Schultz M, Larsen CS, Kreutz K, Larsen CS. 2001. Disease in Spanish Florida: Microscopy of porotic hyperostosis and inferences about health. In: Bioarchaeology of Spanish Florida: The Impact of Colonialism. Gainesville, Florida: University Press of Florida. p 207–225.

Schurr MR. 1992. Isotopic and mortuary variability in a Middle Mississippian population. Am Antiq 57:300–320.

Schurr MR, Powell ML. 2005. The role of changing childhood diets in the prehistoric evolution of food production: an isotopic assessment. Am J Phys Anthropol 126:278–294.

Sciulli PW. 2002. A Survey of Pathological Conditions in the Southwestern Pennsylvania Monongahela. Archaeol East N Am 30:39–57.

Sciulli PW, Giesen MJ, Paine RR. 1996. Paleodemogaphy of the Pearson Complex (33SA9) Eiden Phase Cemetery. Archaeol East N Am 24.

Seeber P, Shander A. 2012. Basics of Blood Management. Wiley.

Semba RD, Bloem MW. 2002. The anemia of vitamin A deficiency: epidemiology and pathogenesis. Eur J Clin Nutr 56:271–281.

Sianto L, Reinhard KJ, Chame M, Chaves S, Mendonça S, Gonçalves ML, Fernandes A, Ferreira LF, Araújo A. 2005. The finding of Echinostoma (Trematoda: Digenea) and hookworm eggs in coprolites collected from a Brazilian mummified body dated 600-1,200 years before present. J Parasitol 91:972–975.

Sohn W-M, Chai J-Y, Yong T-S, Eom KS, Yoon C-H, Sinuon M, Socheat D, Lee S-H. 2011. Echinostoma revolutum infection in children, Pursat Province, Cambodia. Emerg Infect Dis 17:117–119.

Stabler SP, Allen RH. 2004. Vitamin B12 deficiency as a worldwide problem. Annu Rev Nutr 24:299–326.

Steckel RH, Sciulli PW, Rose JC. 2002. A health index from skeletal remains. In: Steckel RH, Rose JC, editors. The Backbone of History: Health and Nutrition in the Western Hemisphere. New York: Cambridge University Press. p 61–93.

Stoltzfus RJ, Chwaya HM, Tielsch JM, Schulze KJ, Albonico M, Savioli L. 1997. Epidemiology of iron deficiency anemia in Zanzibari schoolchildren: the importance of hookworms. Am J Clin Nutr 65:153–159.

Stoltzfus RJ, Chway HM, Montresor A, Tielsch JM, Jape JK, Albonico M, Savioli L. 2004. Low dose daily iron supplementation improves iron status and appetite but not anemia, whereas quarterly anthelminthic treatment improves growth, appetite and anemia in Zanzibari preschool children. J Nutr 134:348–356.

Stone AC, Stoneking M. 1999. Analysis of ancient DNA from a prehistoric Amerindian cemetery. Philos Trans R Soc Lond B Biol Sci 354:153–159.

Stothers DM, Abel TJ. 1989. The Position of the“ Pearson Complex” in the Late Prehistory of Ohio. Archaeol East N Am 17:109–141.

Stuart-Macadam P. 1985. Porotic hyperostosis: representative of a childhood condition. Am J Phys Anthropol 66:391.

Stuart-Macadam P. 1987. A radiographic study of porotic hyperostosis. Am J Phys Anthropol 74:511–520.

Stuart-Macadam P. 1992. Porotic hyperostosis: a new perspective. Am J Phys Anthropol 87:39–47.

Tatala S, Svanberg U, Mduma B. 1998. Low dietary iron availability is a major cause of anemia: a nutrition survey in the Lindi District of Tanzania. Am J Clin Nutr 68:171–178.

Thoman EB. 2009. Historic Hookworm Prevalence Rates and Distribution in the Southeastern United States: Selected Findings of the Reckefeller Sanitary Comission for the Eradication of Hookworm. Rockefeller Arch Cent Res Rep Online:1–21.

Tieszen LL, Fagre T. 1993. Effect of diet quality and composition on the isotopic composition of respiratory CO2, bone collagen, bioapatite, and soft tissues. In: Prehistoric Human Bone. Springer. p 121–155.

Trimble CC. 1996. Palaeodiet in Virgina and North Carolina as determined by stable isotope analysis of skeletal remains.

Ubelaker DH. 1974. Reconstruction of demographic profiles from ossuary skeletal samples: a case study from the Tidewater Potomac. Washington DC: Smithsonian Institution Press.

Ucko PJ. 1969. Ethnography and archaeological interpretation of funerary remains. World Archaeol 1:262–280.

van der Merwe N, Vogel JC. 1978. Carbon-13 content of human collagen as a measure of prehistoric diet in woodland North America. Nature 276:815–816.

Vasalech C. 2011. Reinterpreting cribra orbitalia etiology in a coastal North Carolina Algonkian population using computed tomography.

Vogel JR. 2007. Mound versus village: a biocultural investigation of status and health at the Cox Site.

Walker PL, Bathurst RR, Richman R, Gjerdrum T, Andrushko VA. 2009. The causes of porotic hyperostosis and cribra orbitalia: A reappraisal of the iron-deficiency-anemia hypothesis. Am J Phys Anthropol 139:109–125.

Walsh MG. 2013. Forest Fragmentation and Risk of Giardiasis in New York State. EcoHealth 10:405–414.

Wapler U, Crubezy E, Schultz M. 2004. Is cribra orbitalia synonymous with anemia? Analysis and interpretation of cranial pathology in Sudan. Am J Phys Anthropol 123:333–339.

Williams KD. 2005. a geospatial bioarchaeological perspective on behavior, lifestyle, and activity patterns in the eastern woodlands of North America.

Wood JW, Mimer GR, Harpending HC, Weiss KM. 1992. The Osteological Paradox. Curr Anthropol 33:343–370.

Wright LE, Yoder CJ. 2003. Recent progress in bioarchaeology: approaches to the osteological paradox. J Archaeol Res 11:43–70.

Zeiler M. 1999. Modeling our world: the ESRI guide to geodatabase design. Redlands, California: ESRI Press.

APPENDIX A

Maps for each environmental variable at each archaeological site

A1. Soil Drainage

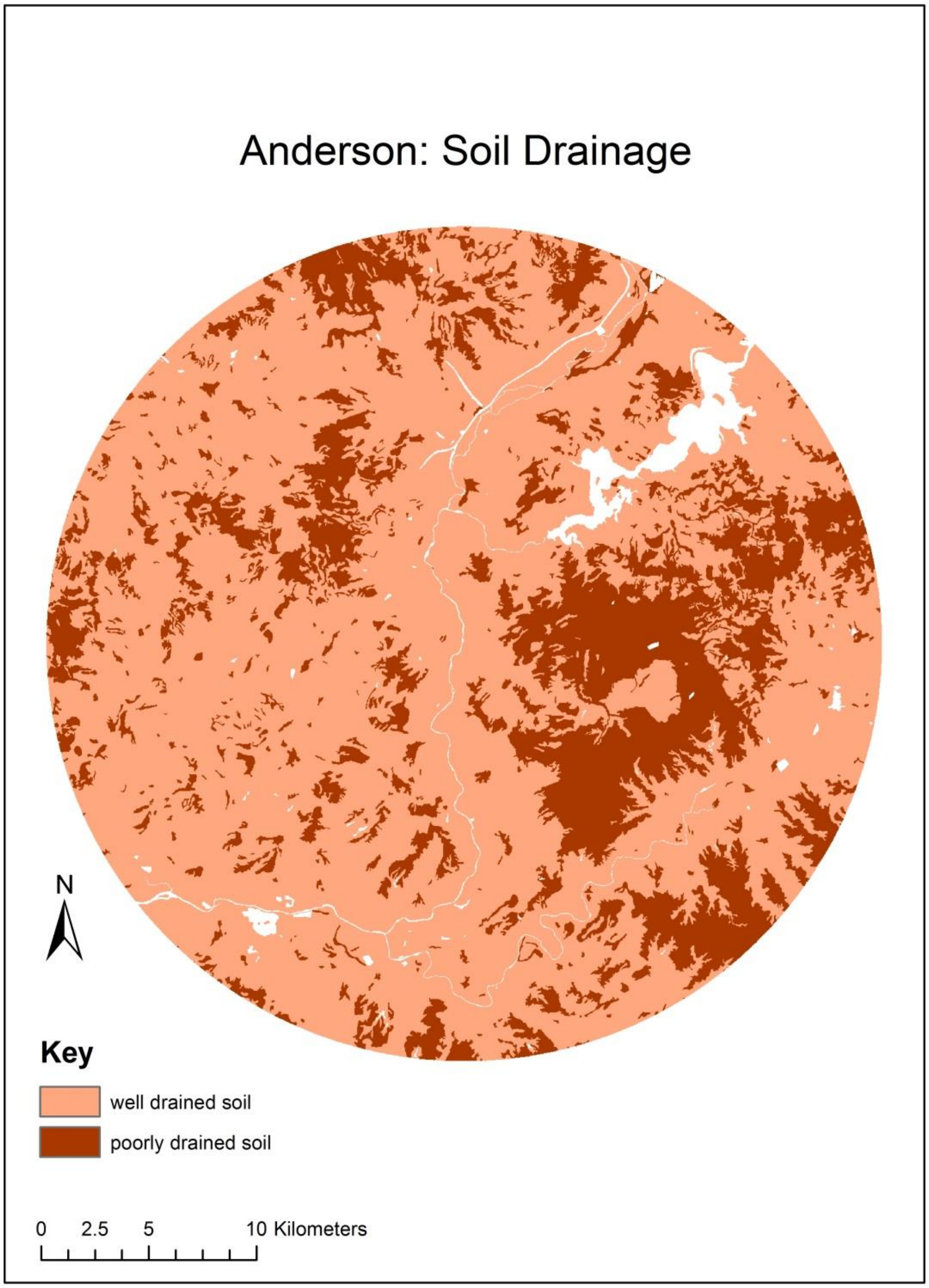

Figure 16. Soil drainage at Anderson.

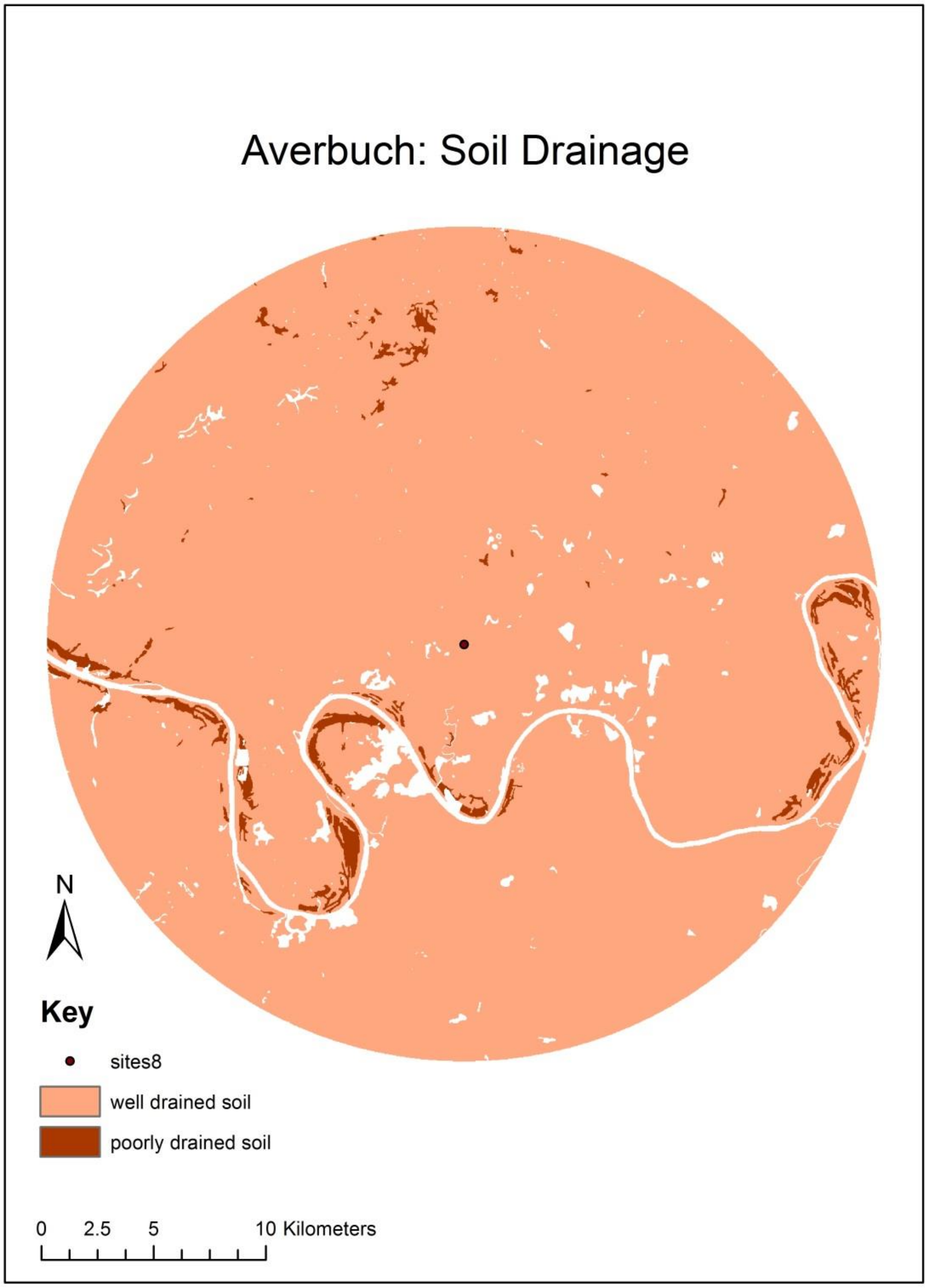

Figure 17. Soil drainage at Averbuch.

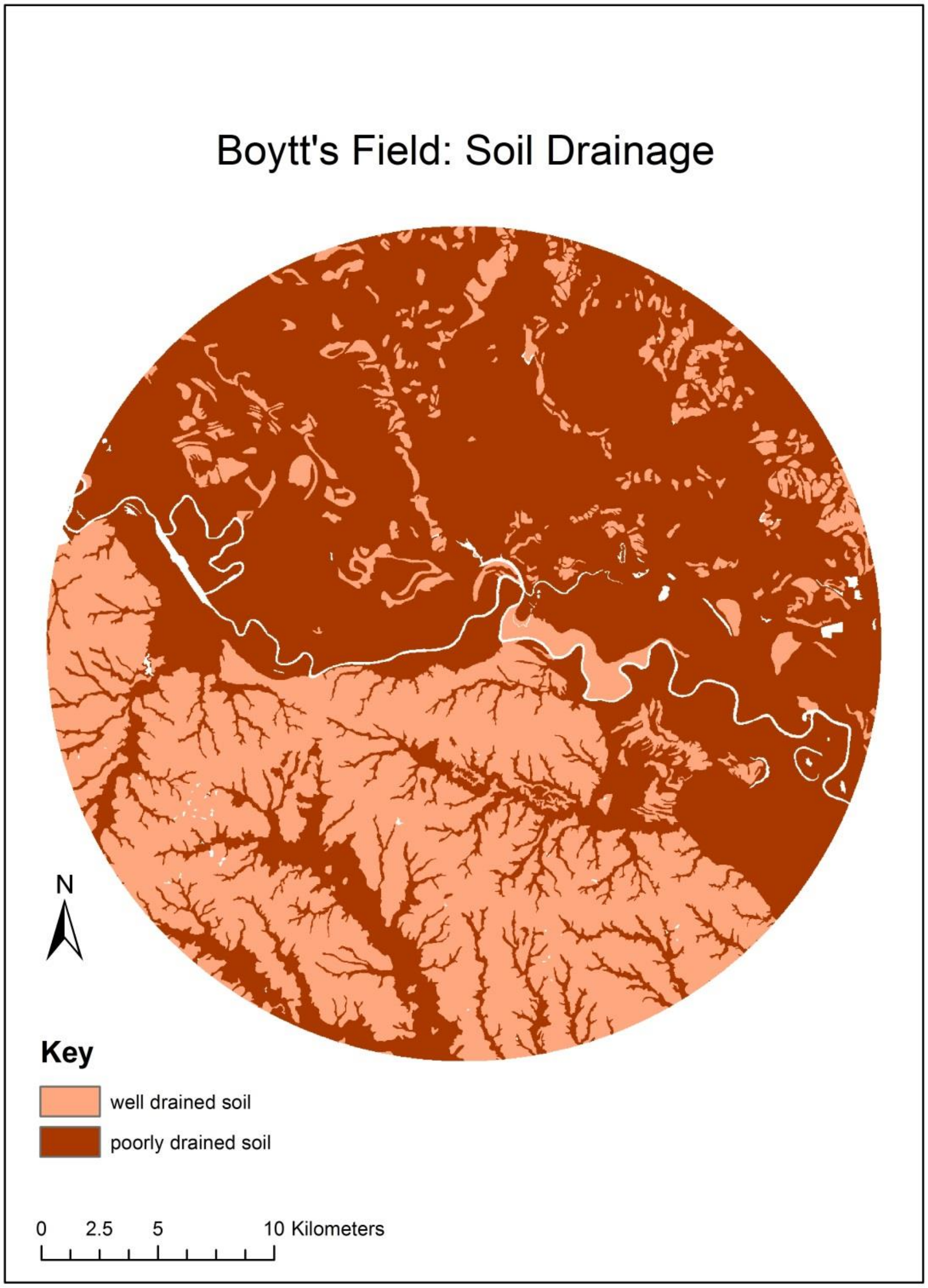

Figure 18. Soil drainage at Boytt's Field.

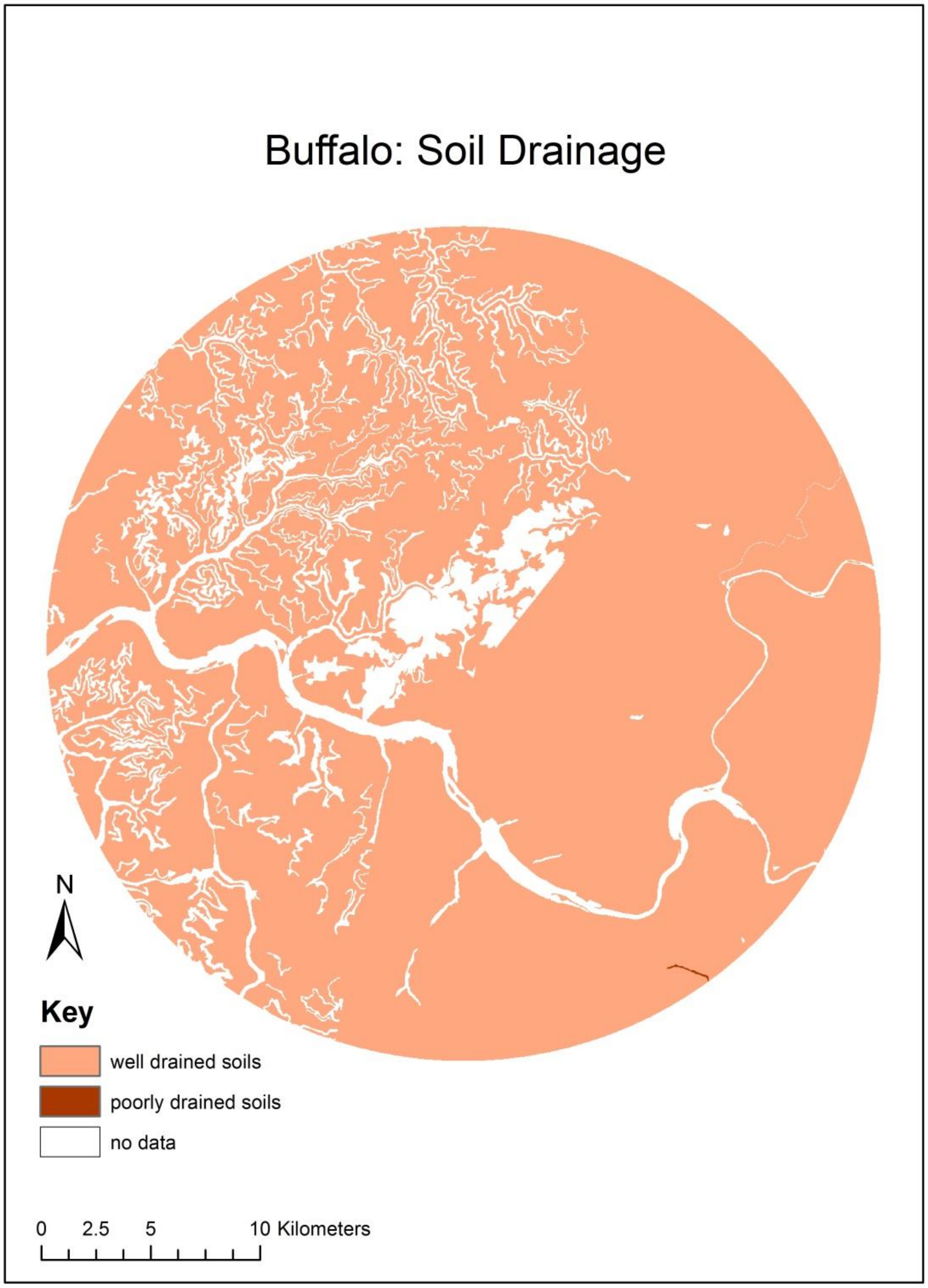

Figure 19. Soil drainage at Buffalo.

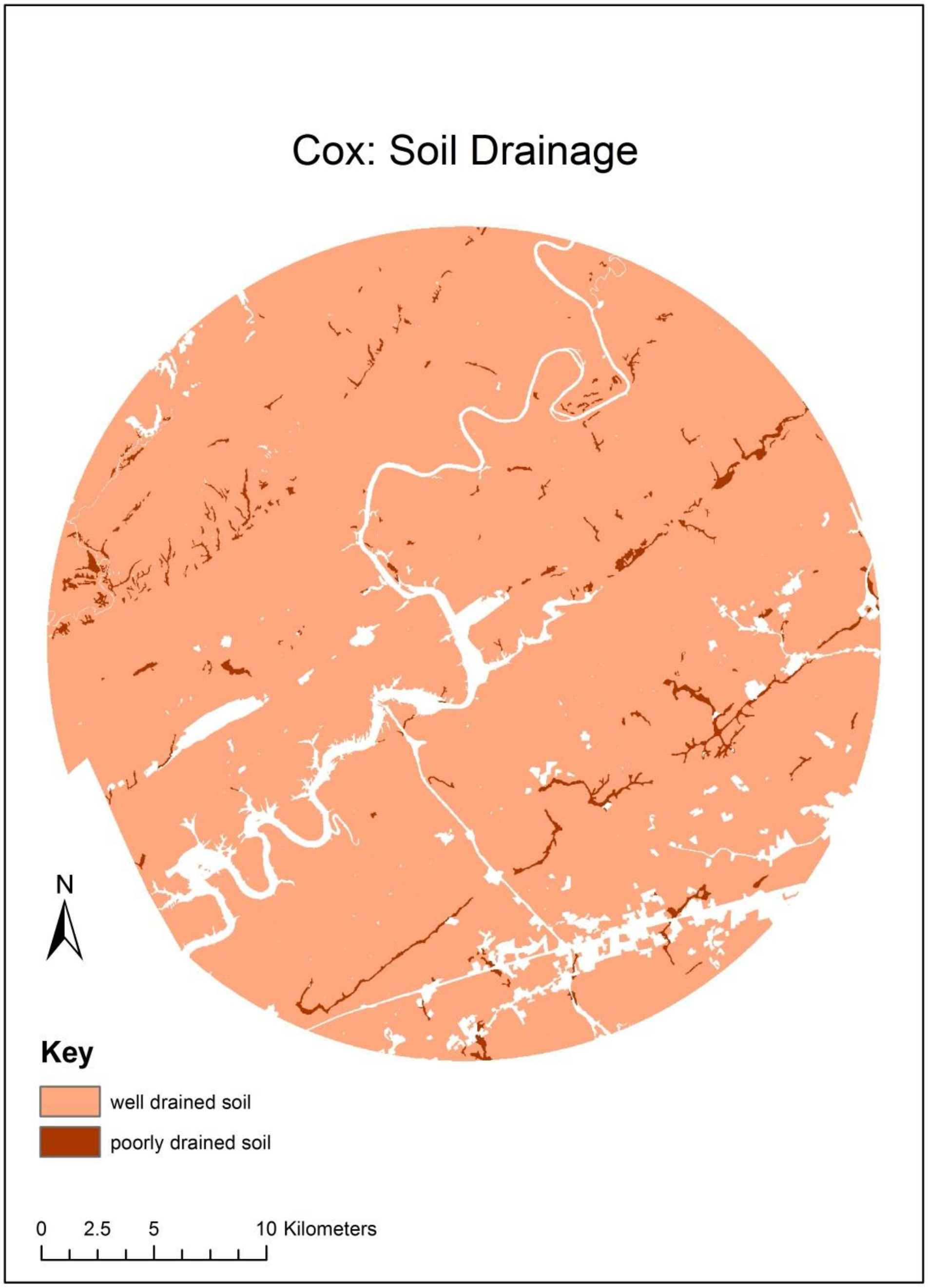

Figure 20. Soil drainage at Cox.

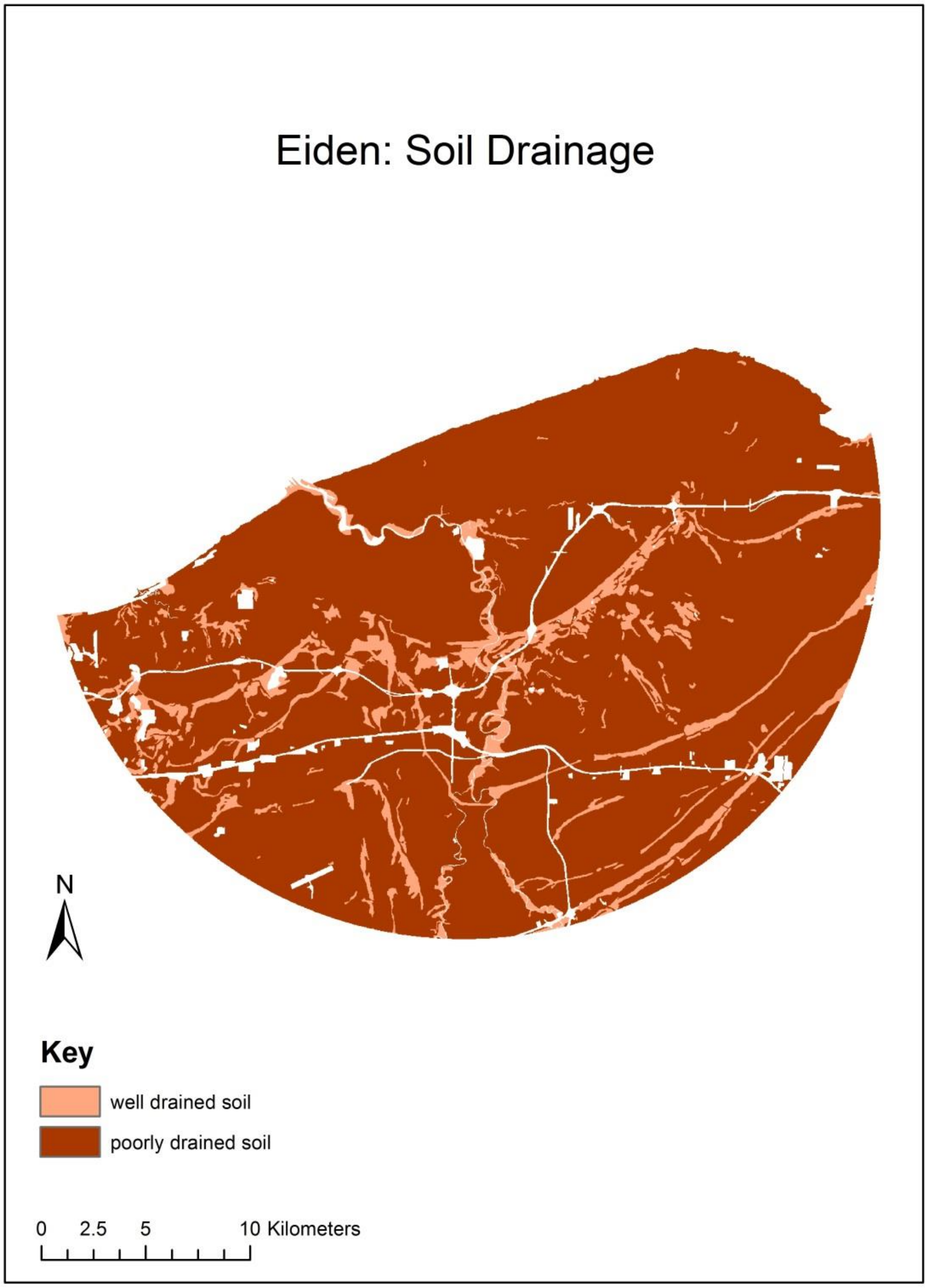

Figure 21. Soil drainage at Eiden.

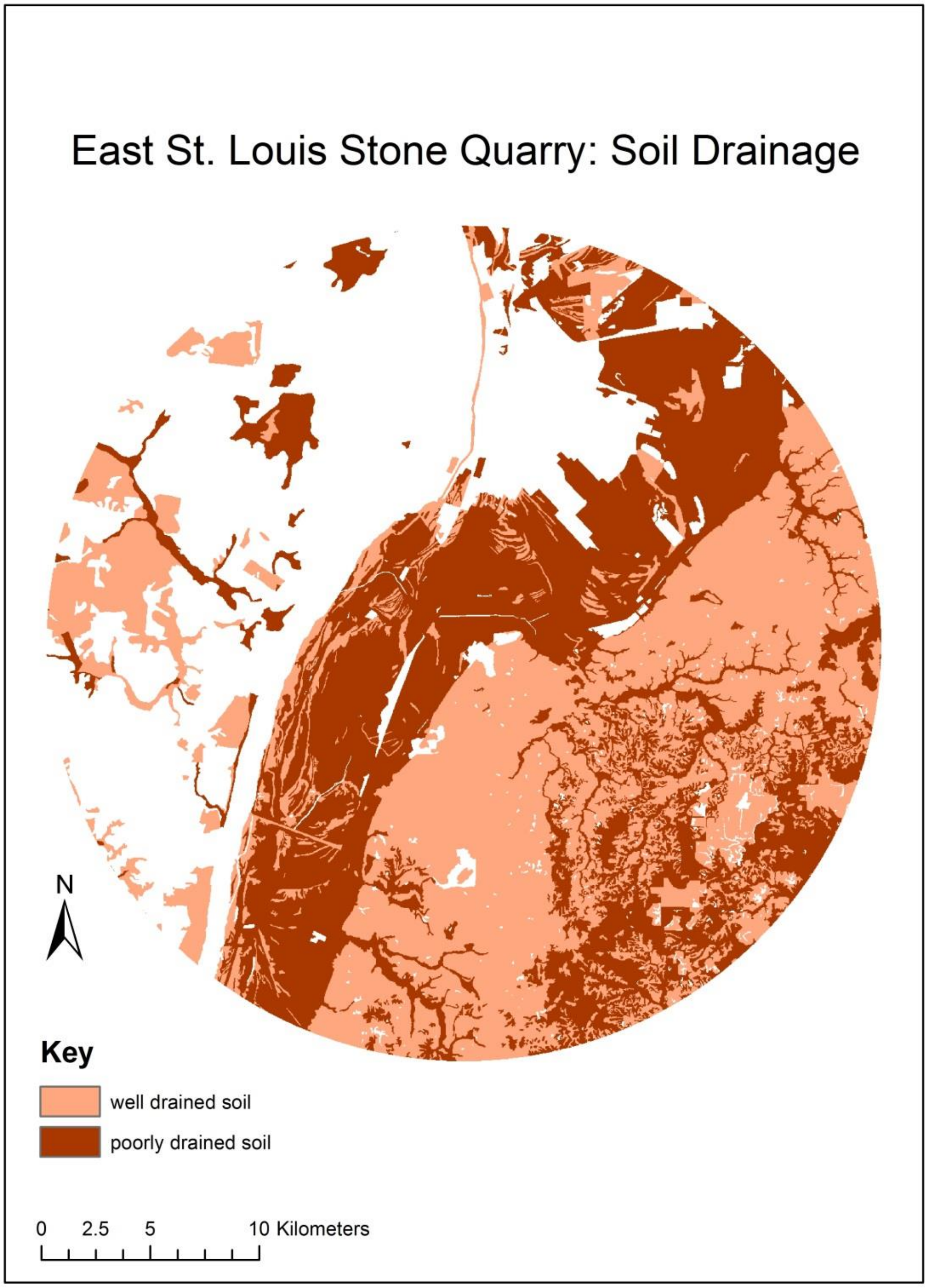

Figure 22. Soil drainage at East St. Louis Stone Quarry.

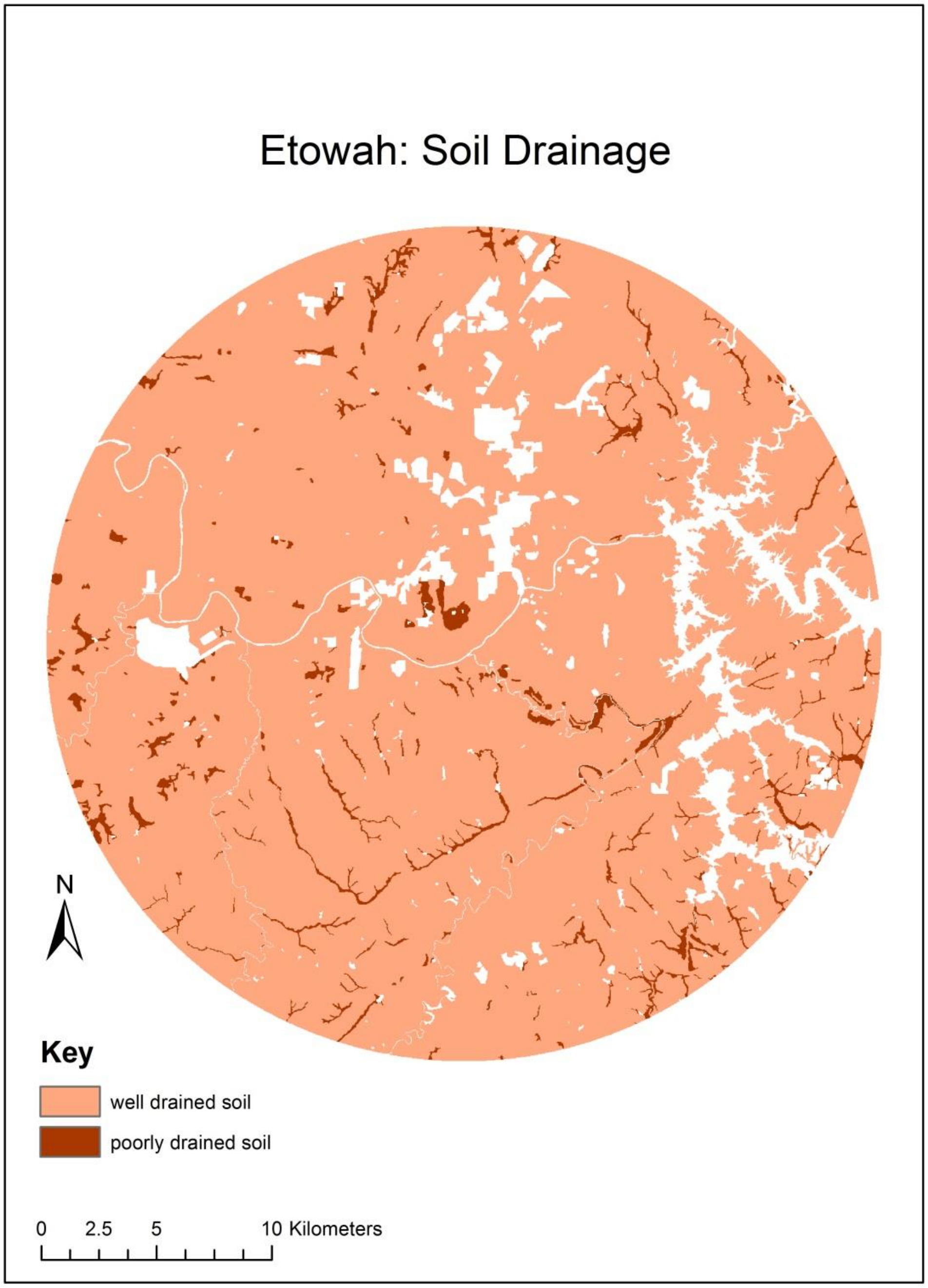

Figure 23. Soil drainage at Etowah.

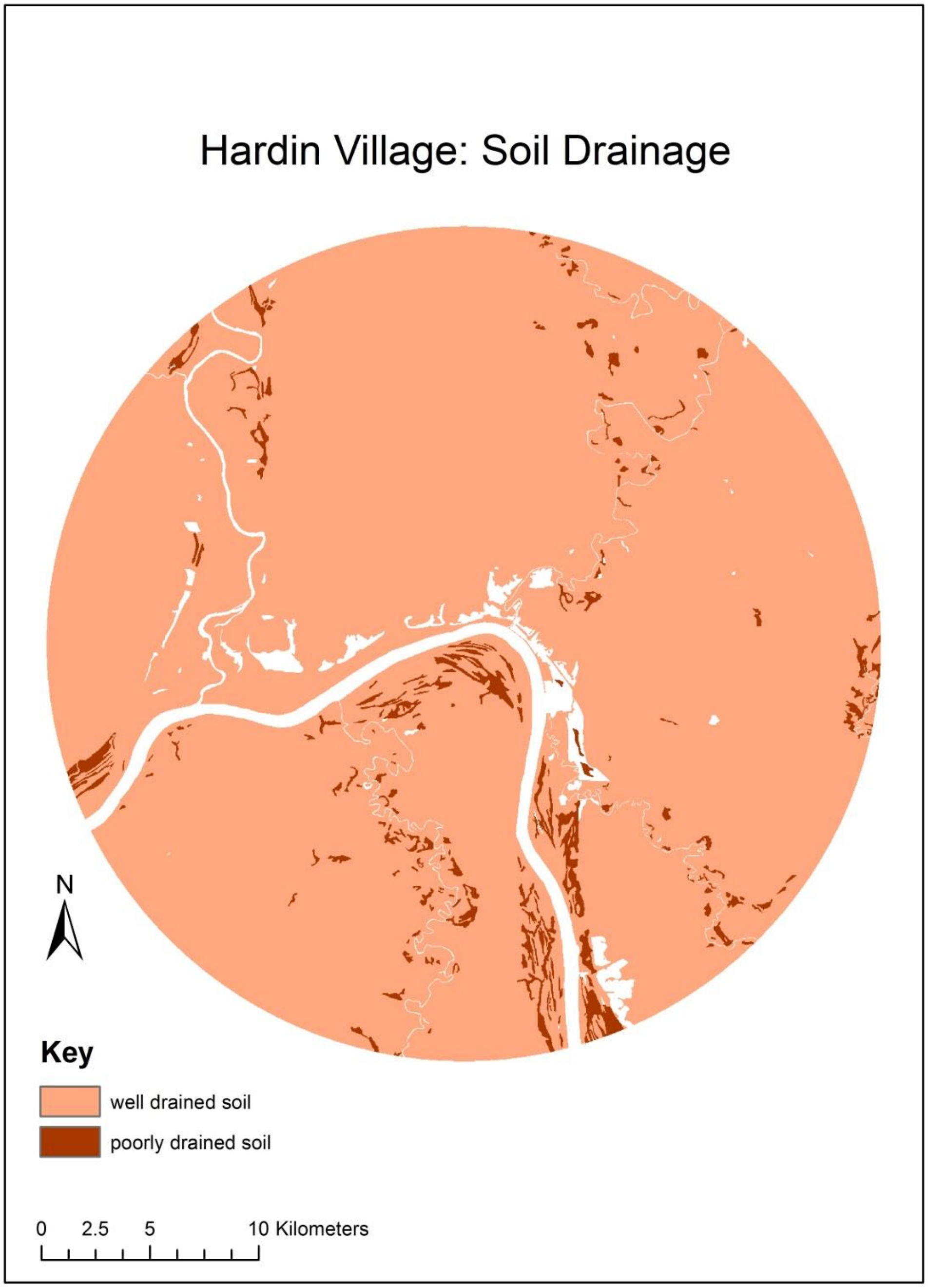

Figure 24. Soil drainage at Hardin Village.

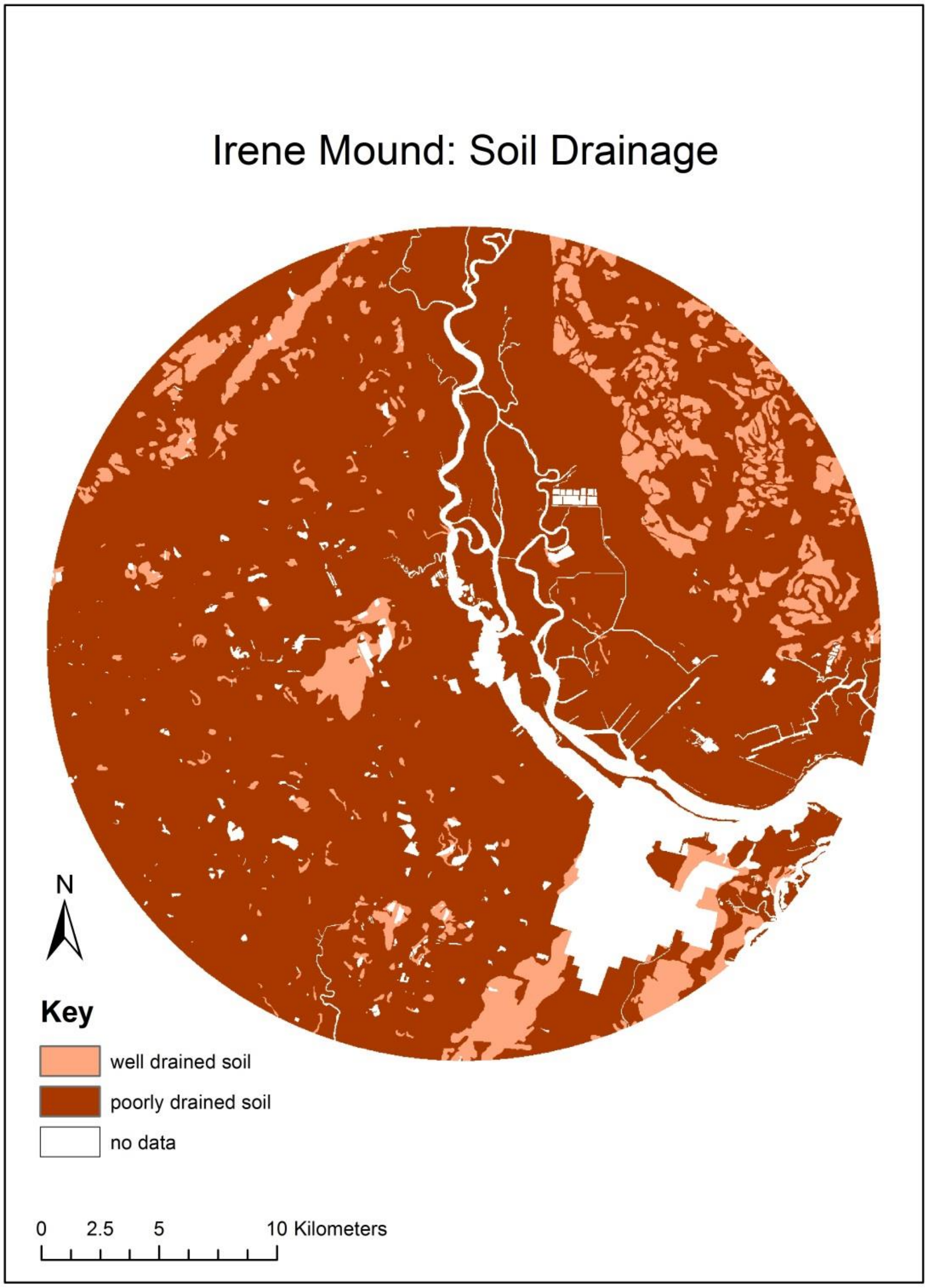

Figure 25. Soil drainage at Irene Mound.

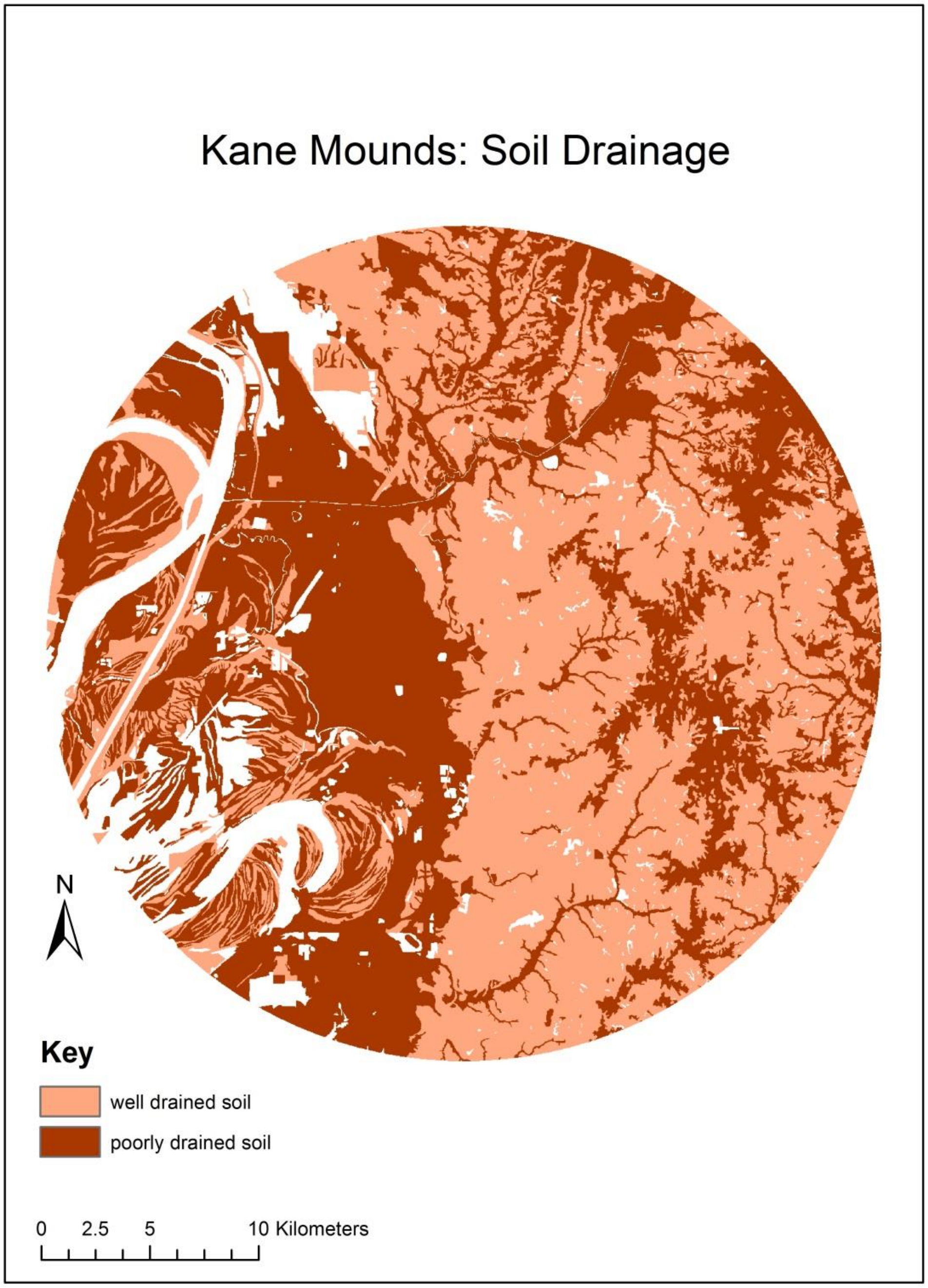

Figure 26. Soil drainage at Kane Mounds.

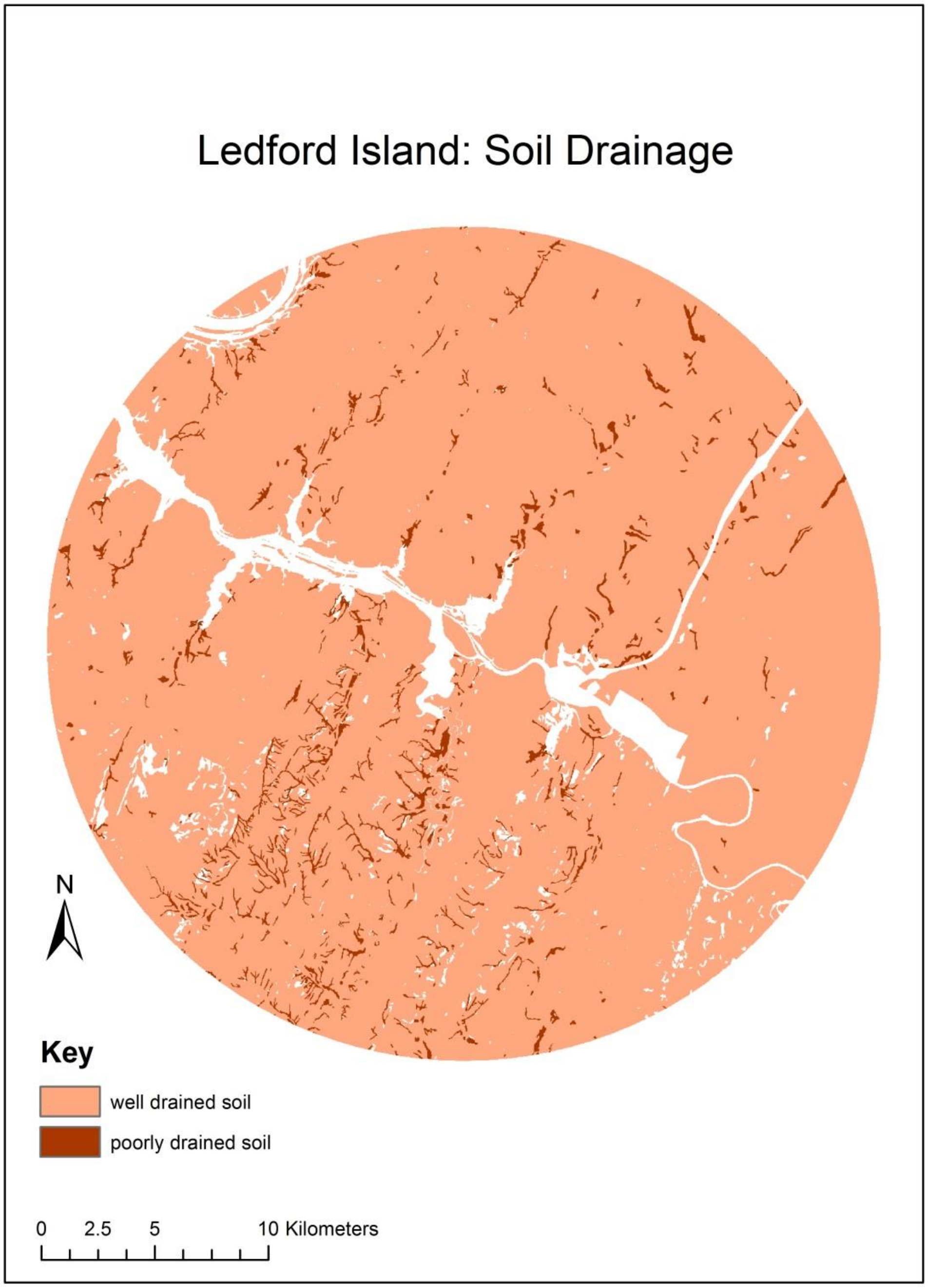

Figure 27. Soil drainage at Ledford Island.

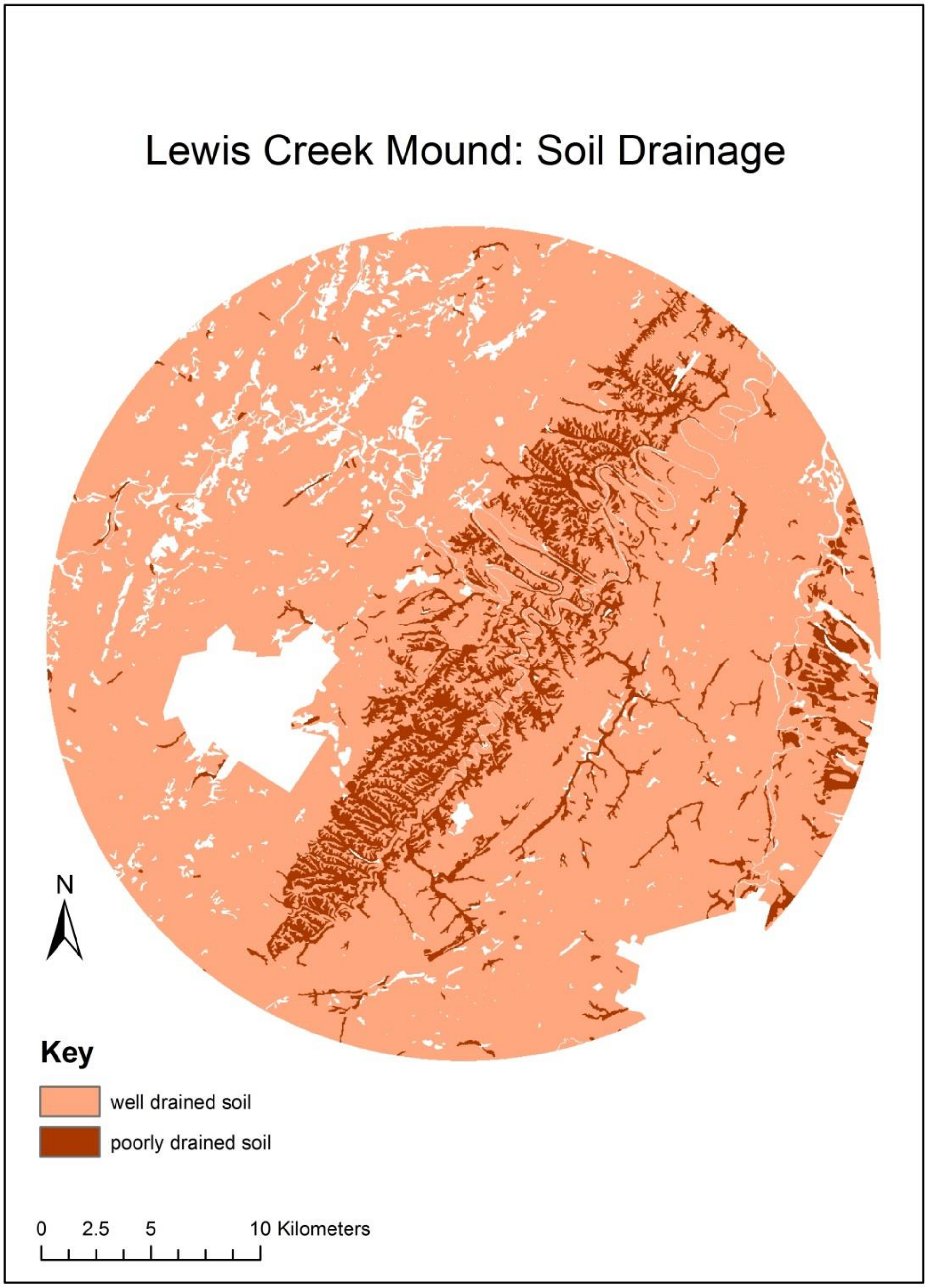

Figure 28. Soil drainage at Lewis Creek Mound.

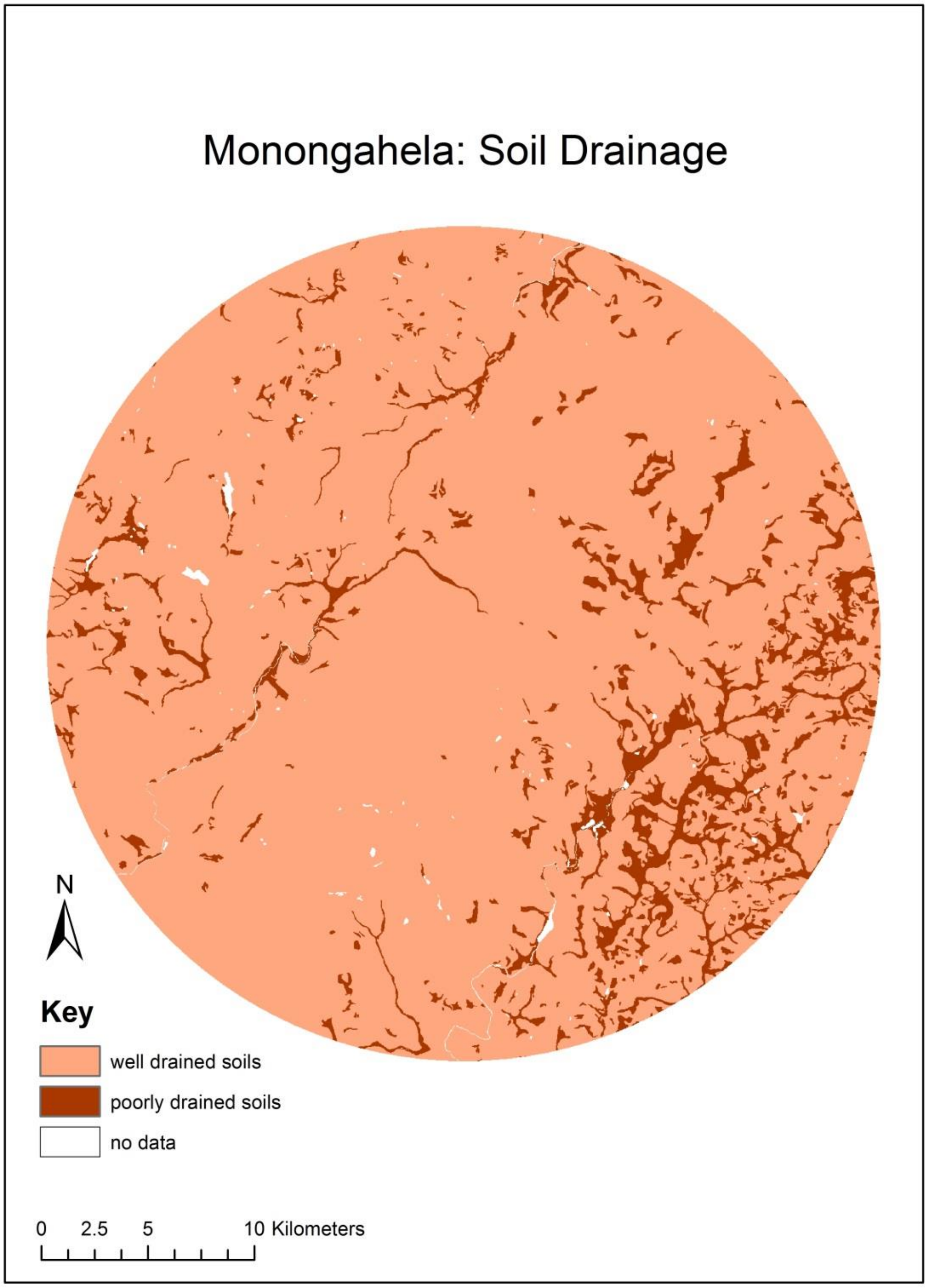

Figure 29. Soil drainage at Monongahela.

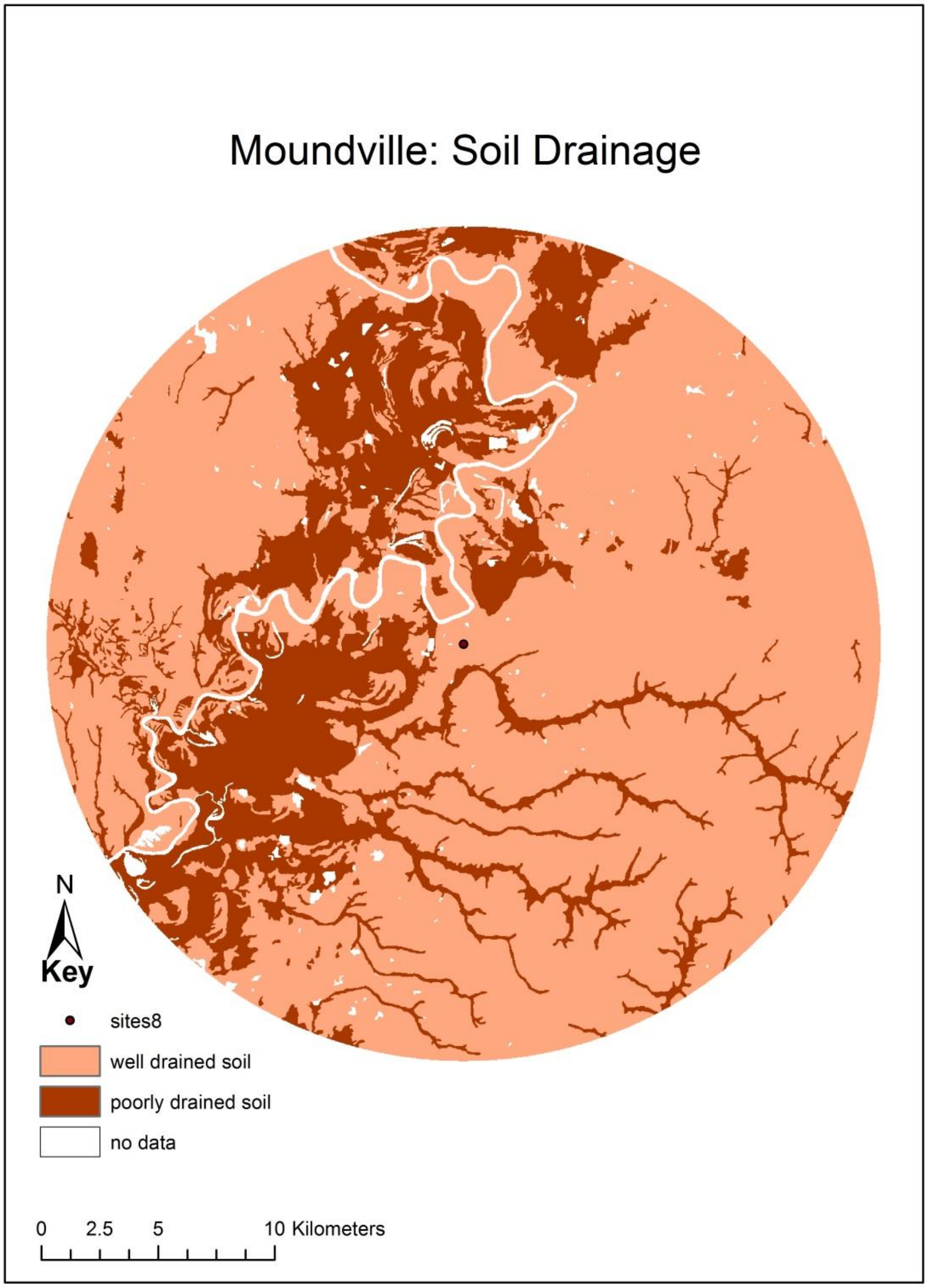

Figure 30. Soil drainage at Moundville.

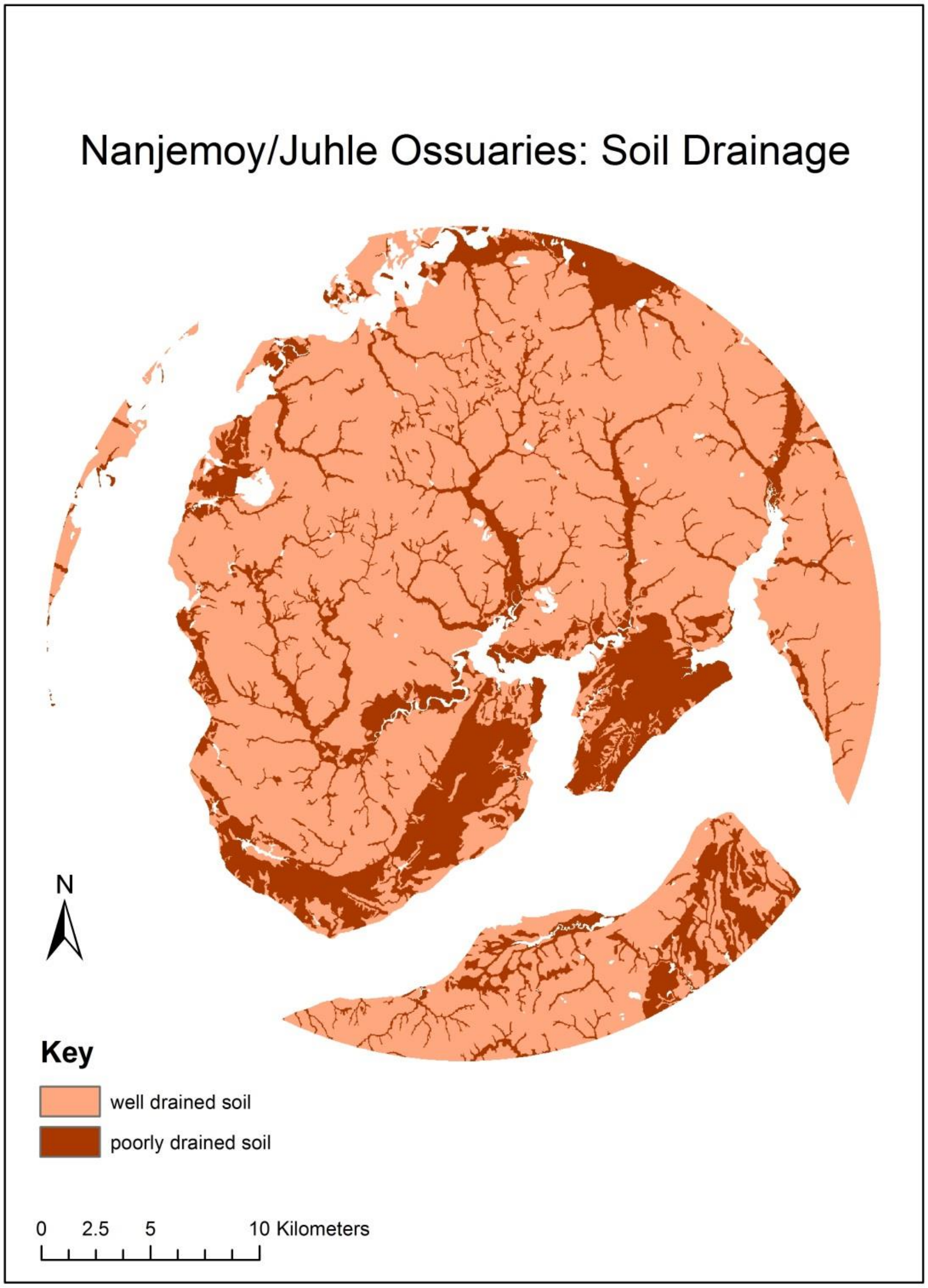

Figure 31. Soil drainage at Juhle Ossuary.

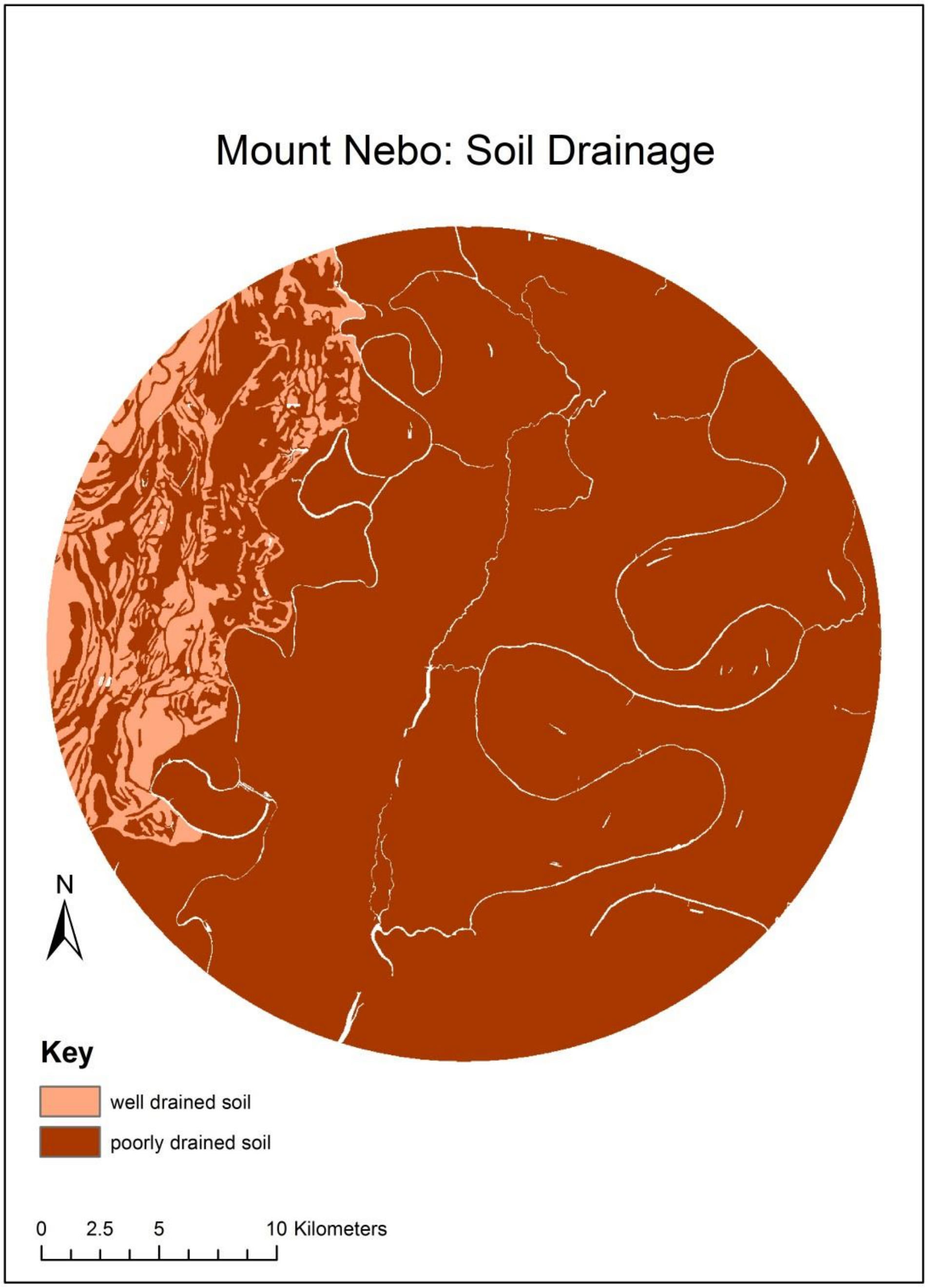

Figure 32. Soil drainage at Mount Nebo.

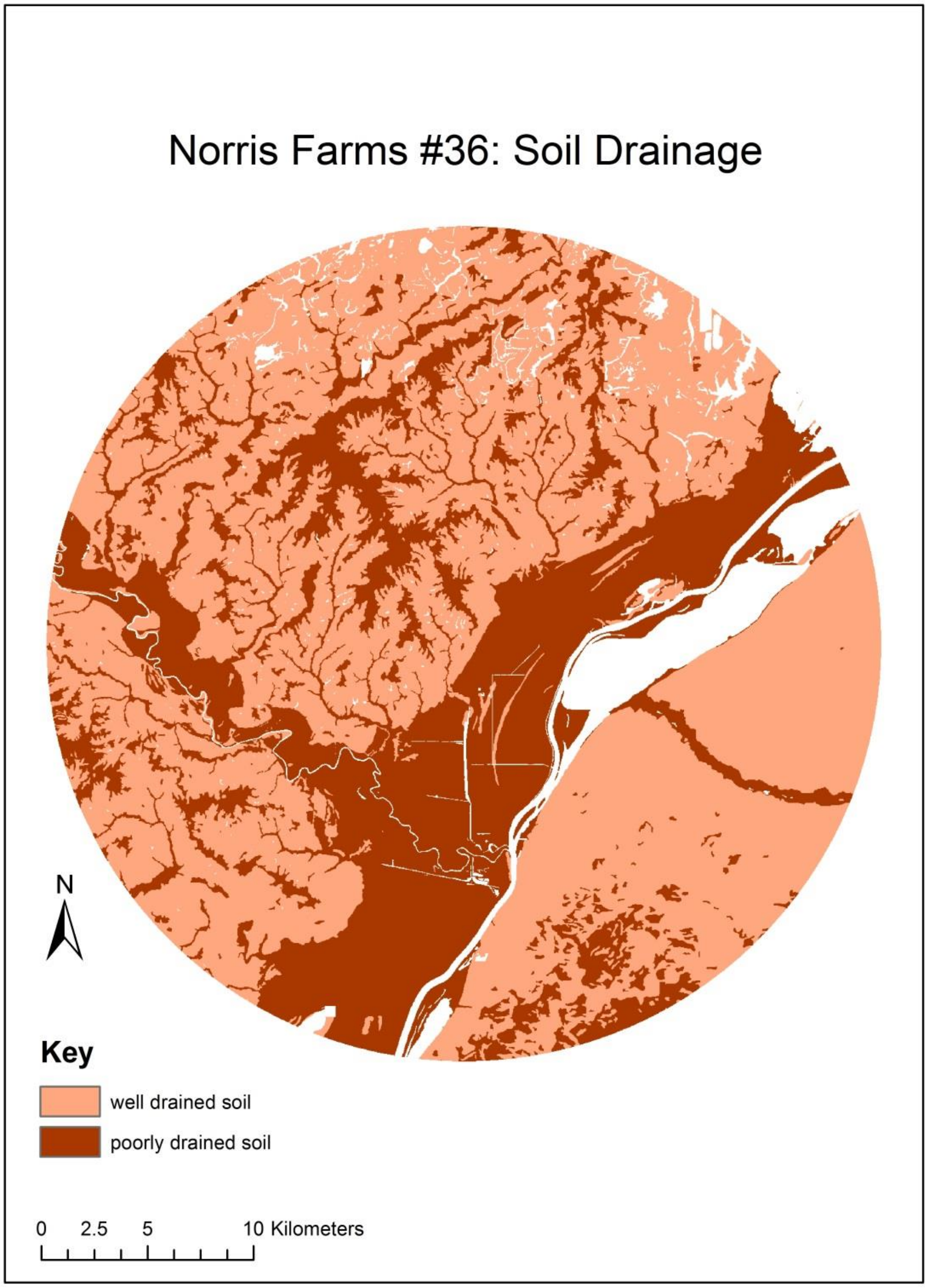

Figure 33. Soil drainage at Norris Farms #36.

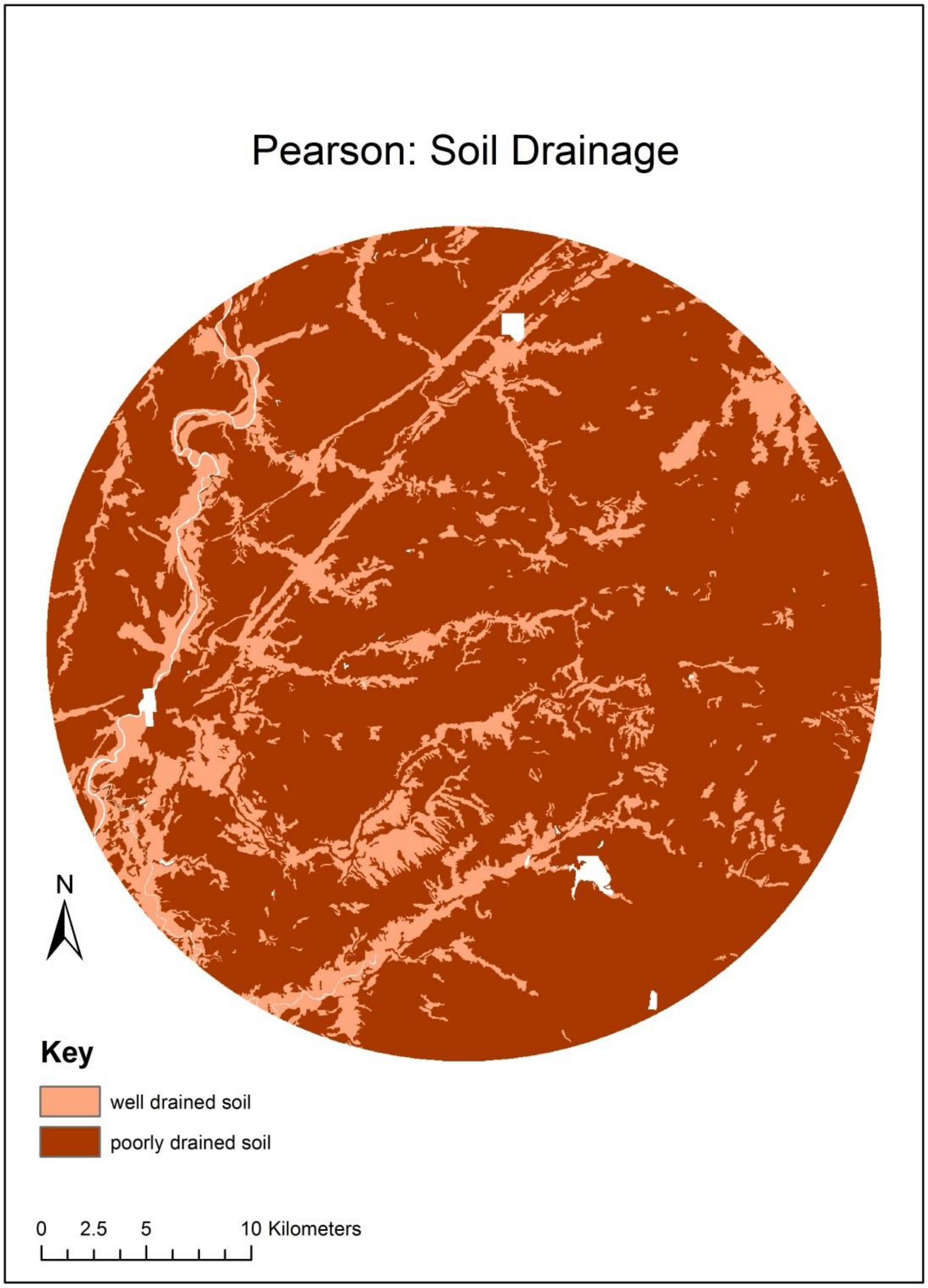

Figure 34. Soil drainage at Pearson.

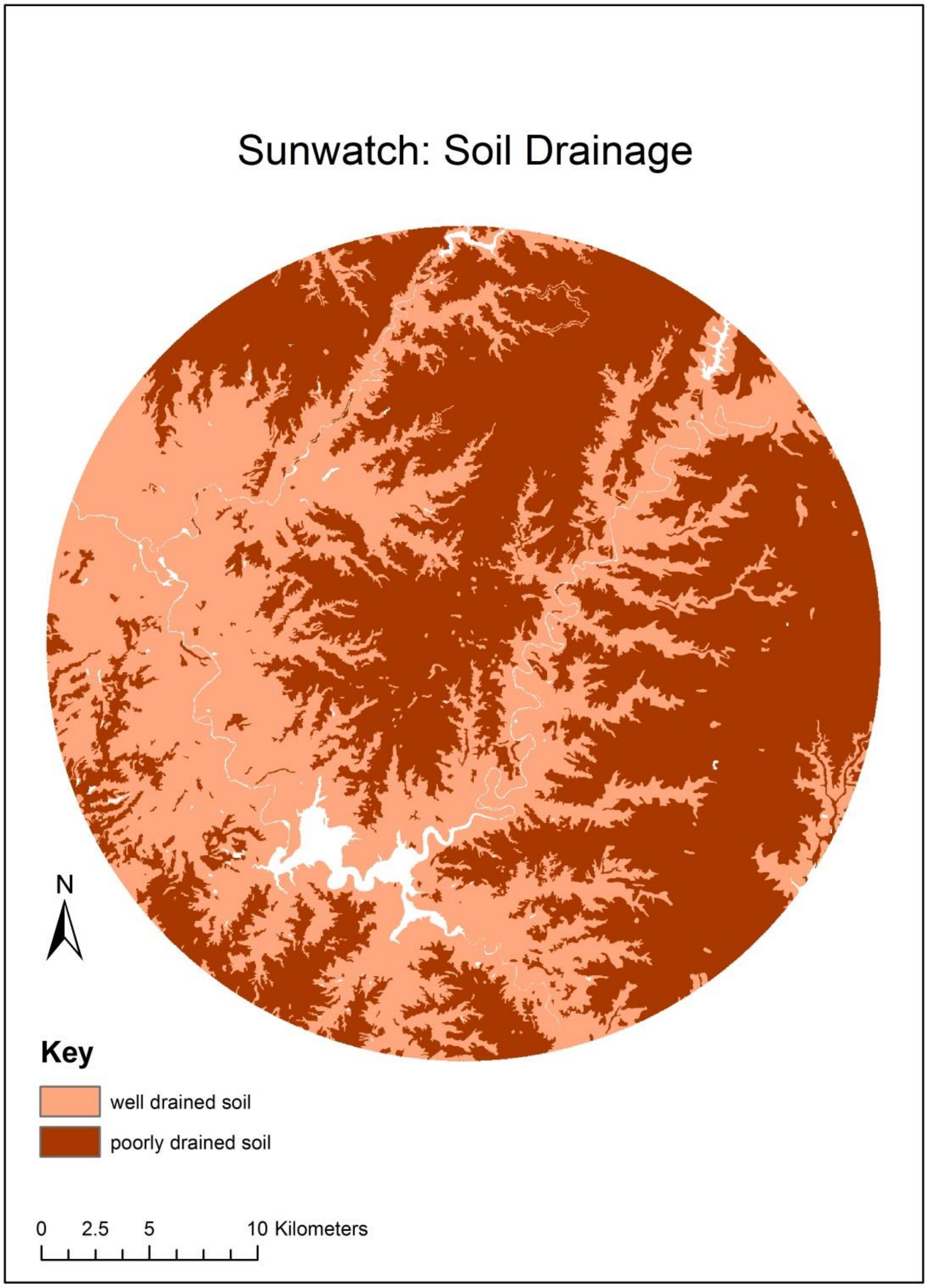

Figure 35. Soil drainage at Sunwatch.

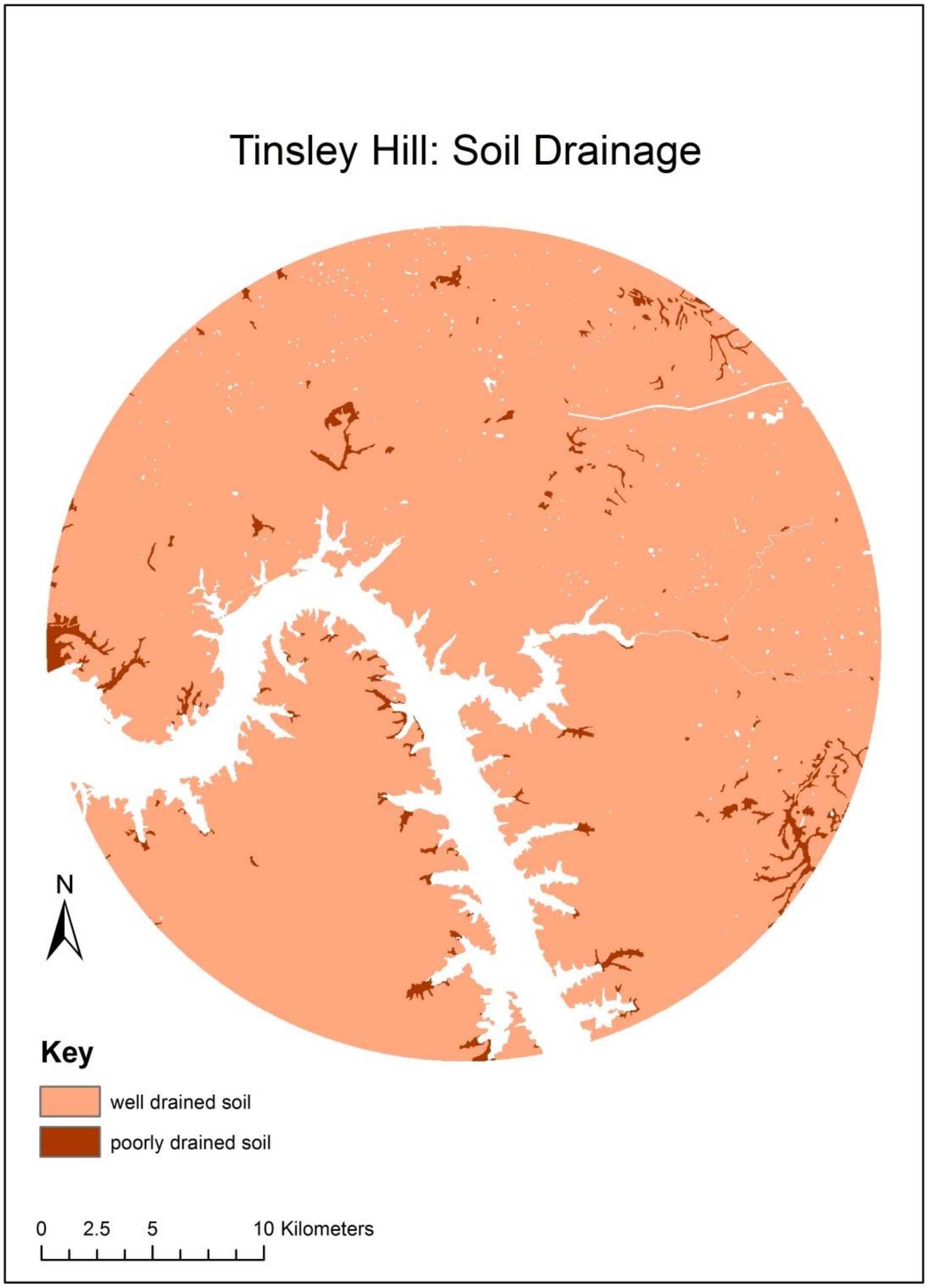

Figure 36. Soil drainage at Tinsley Hill.

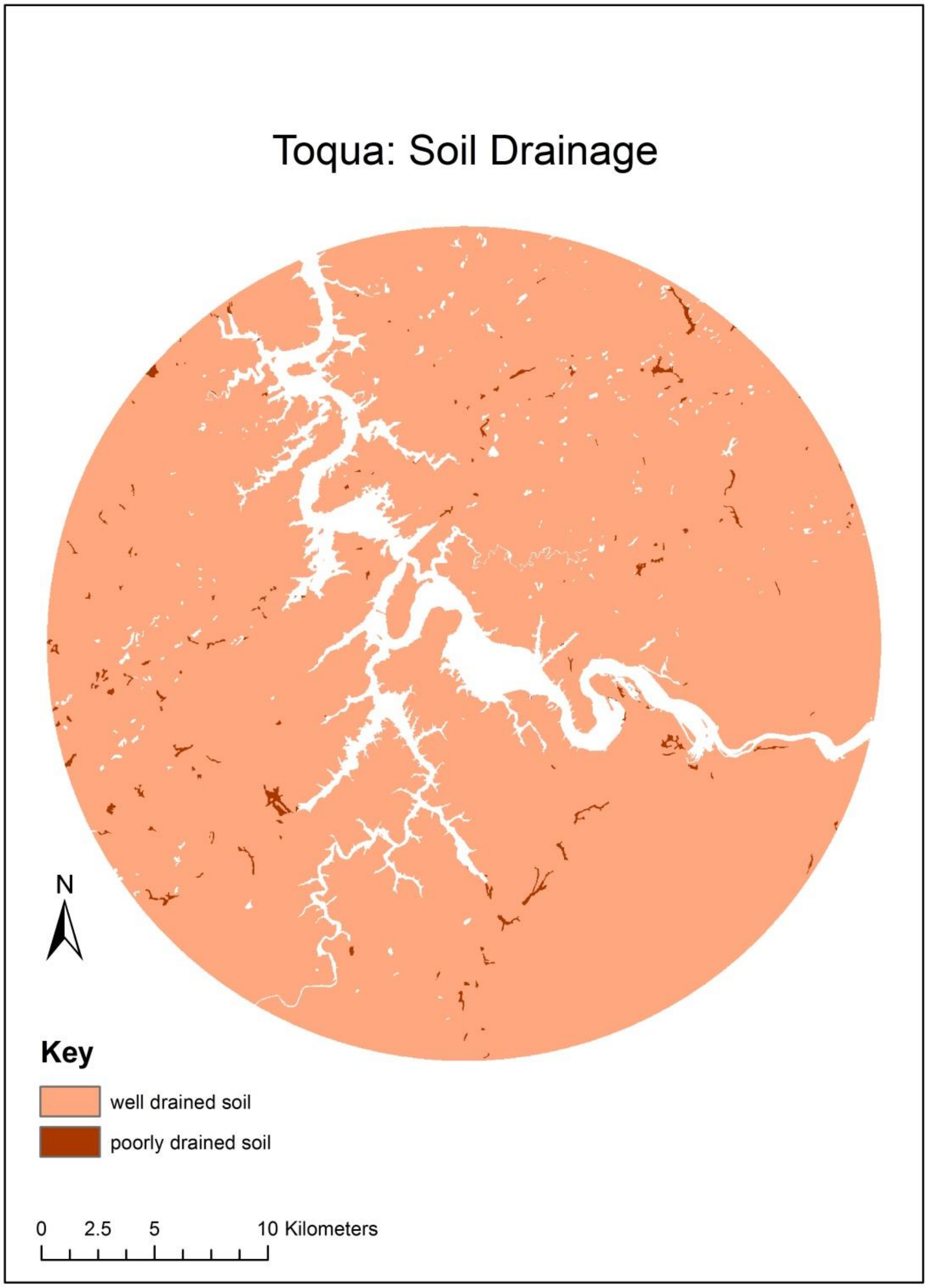

Figure 37. Soil drainage at Toqua.

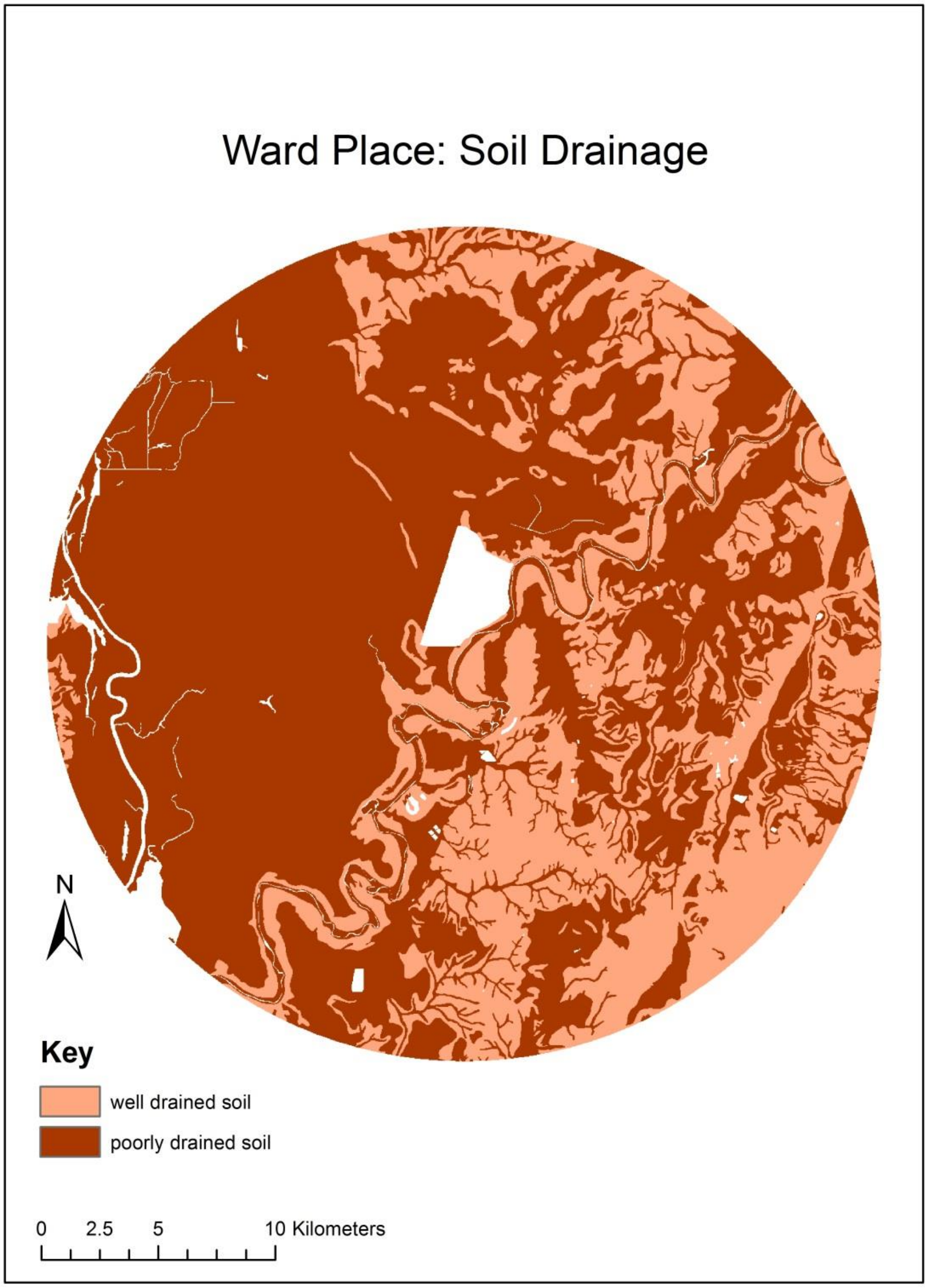

Figure 38. Soil drainage at Ward Place.

A2. Temperature

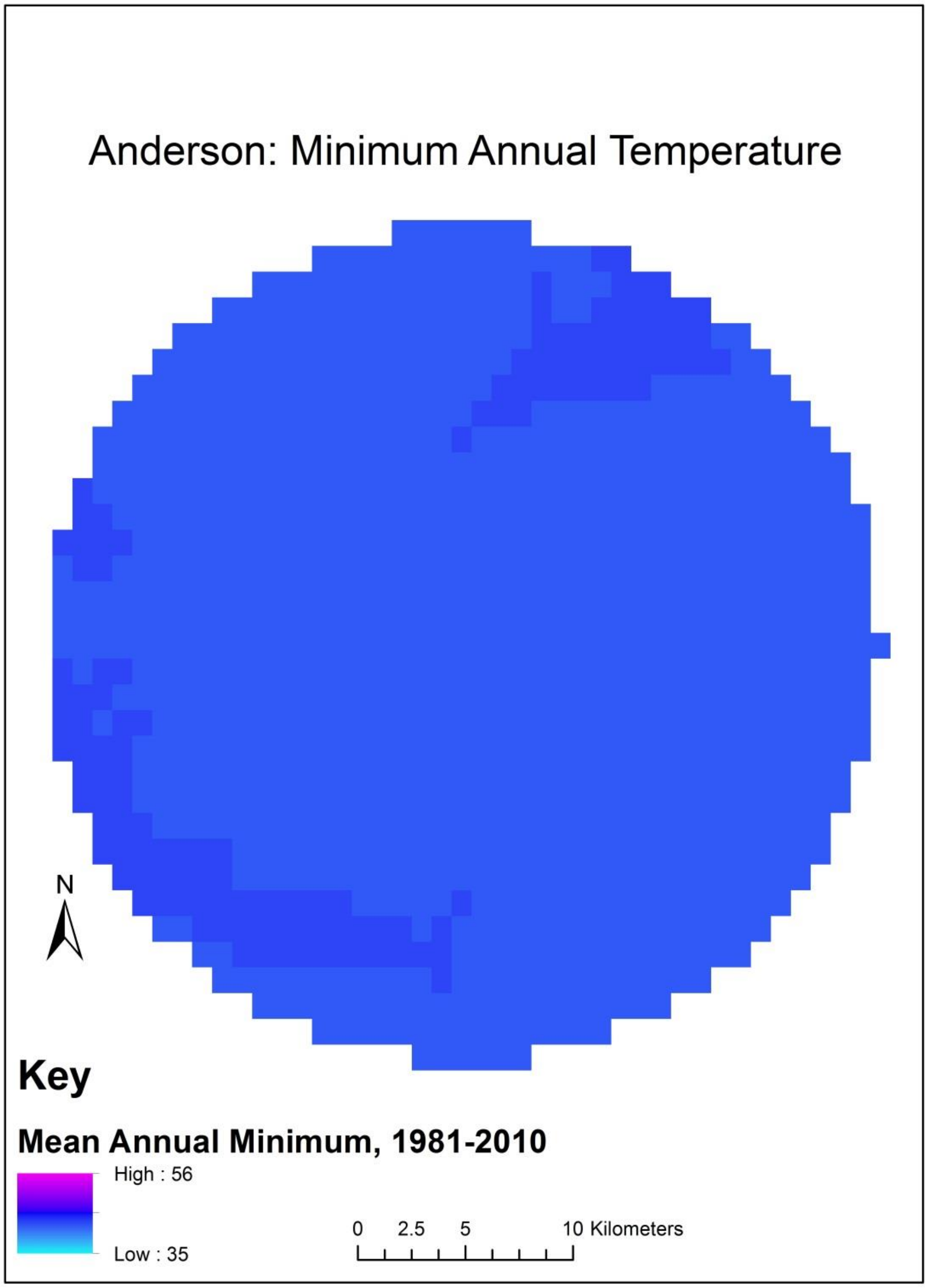

Figure 39. Temperature at Anderson.

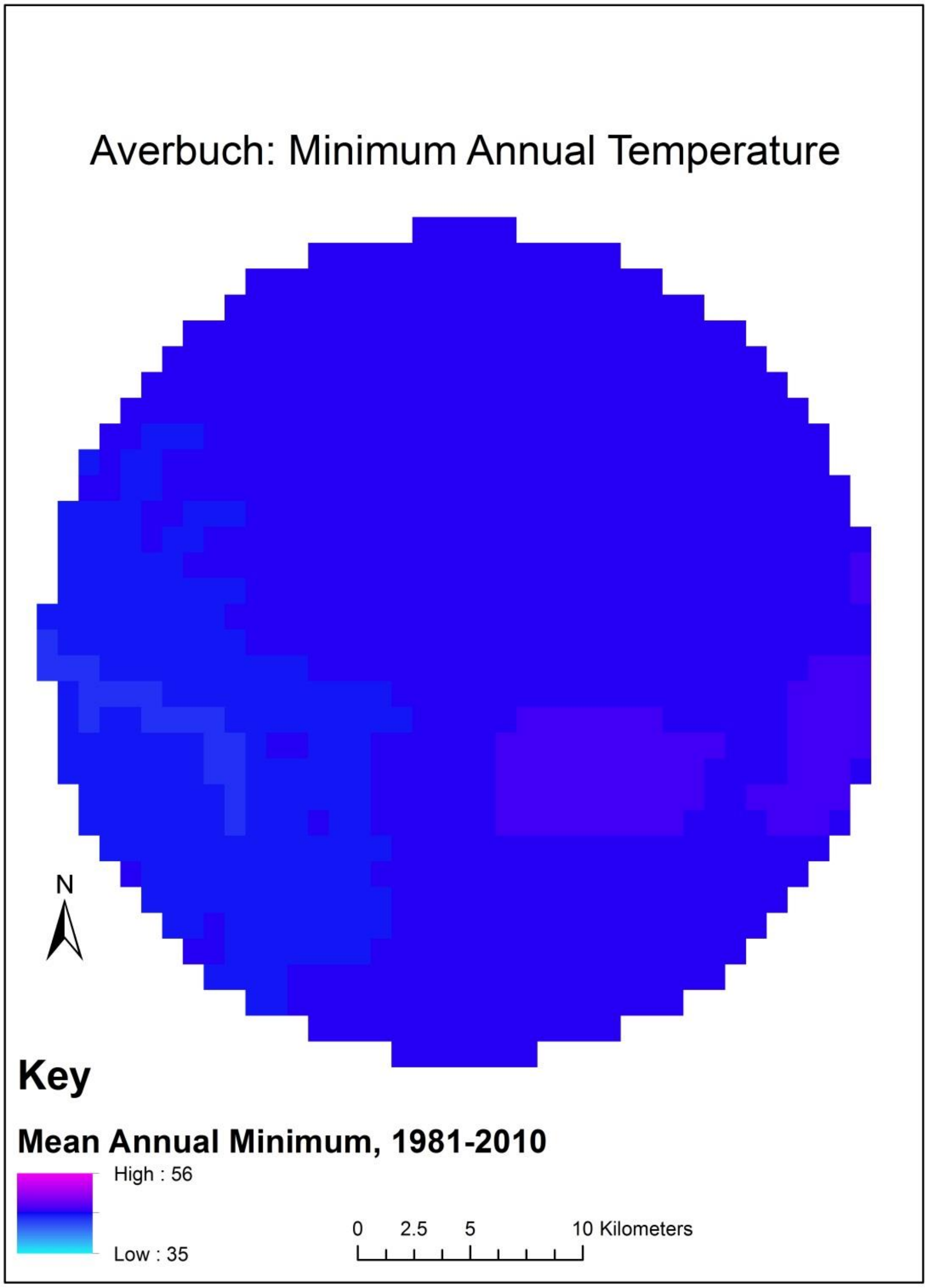

Figure 40. Temperature at Averbuch.

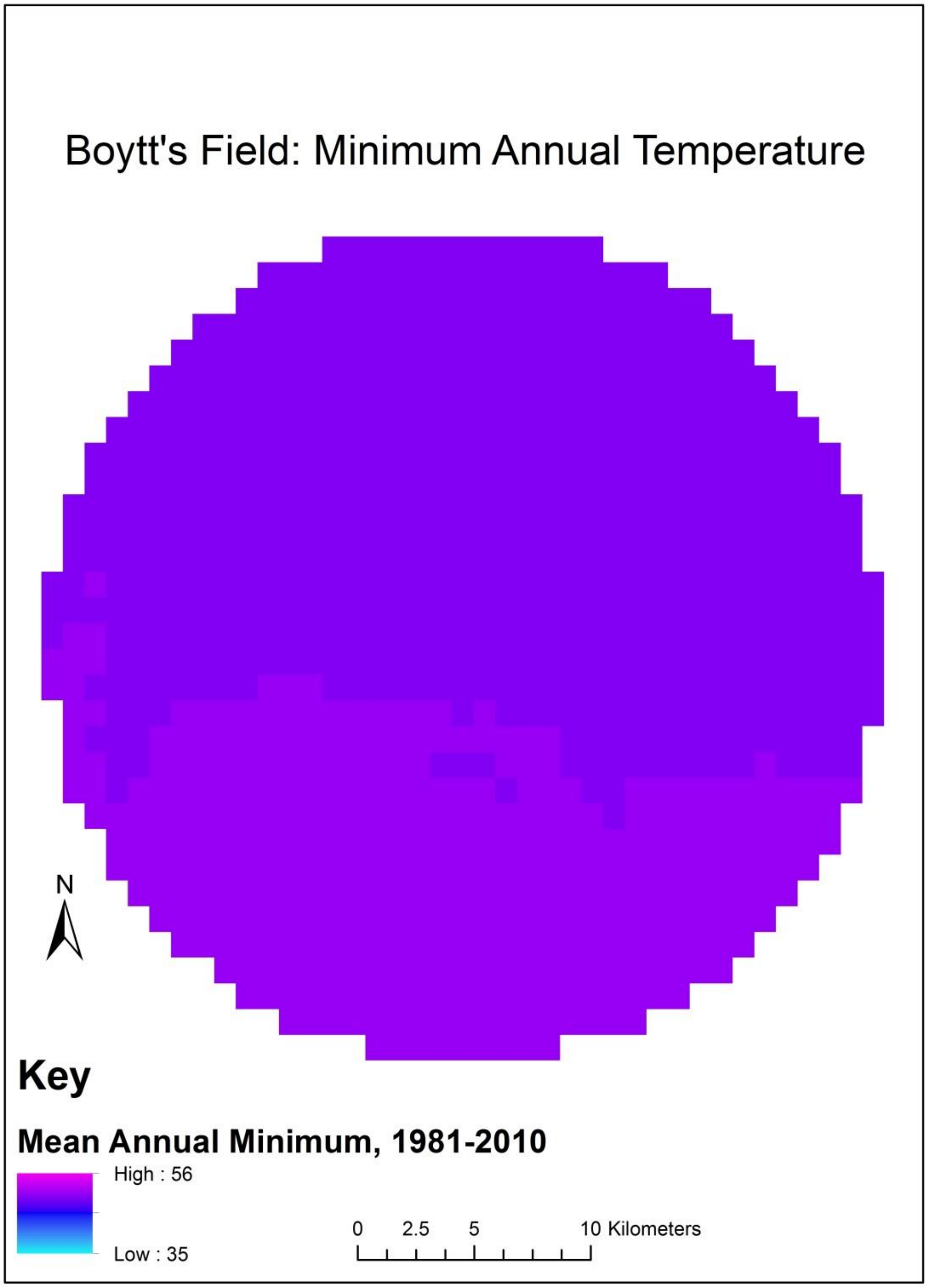

Figure 41. Temperature at Boytt's Field.

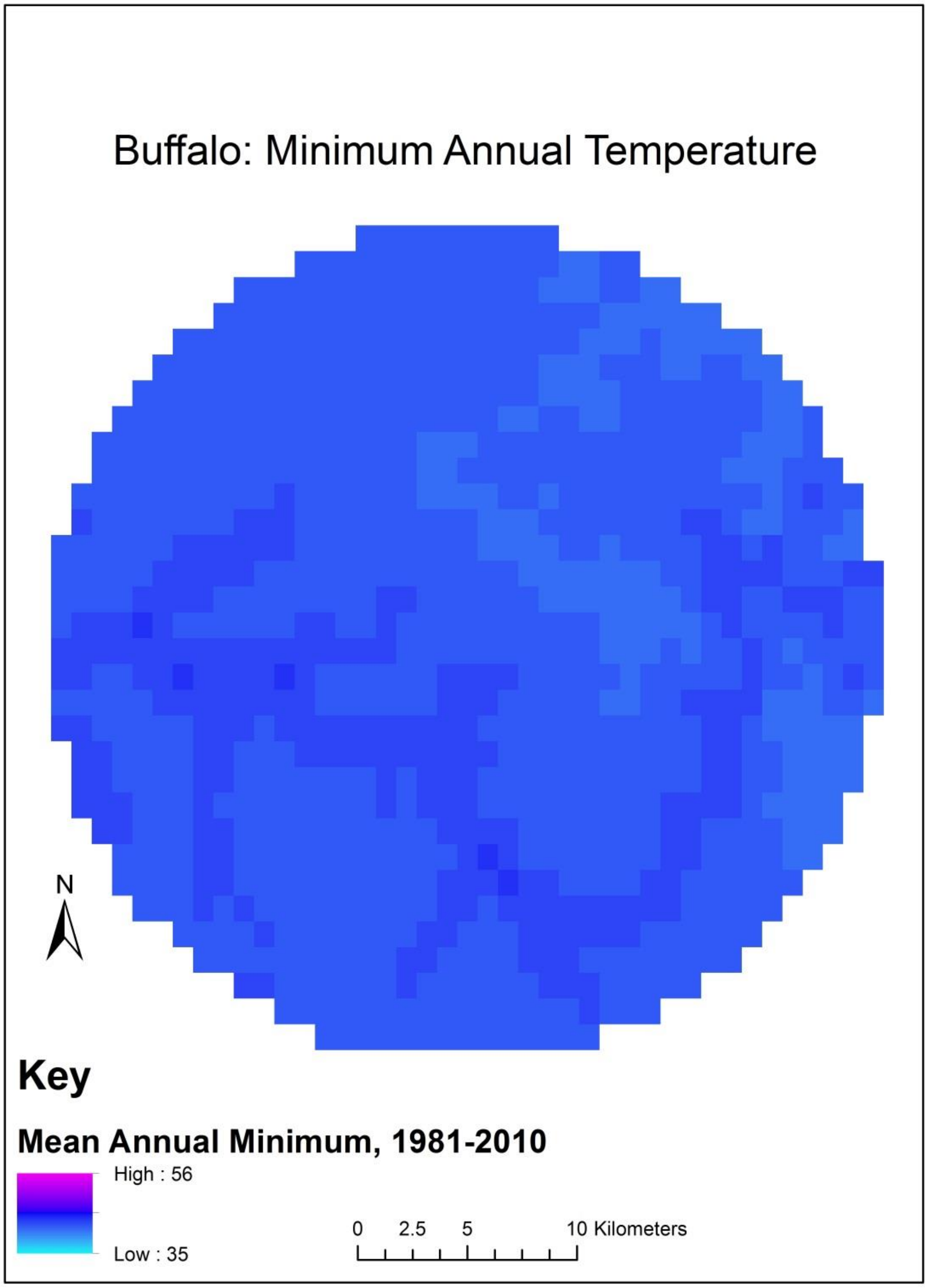

Figure 42. Temperature at Buffalo.

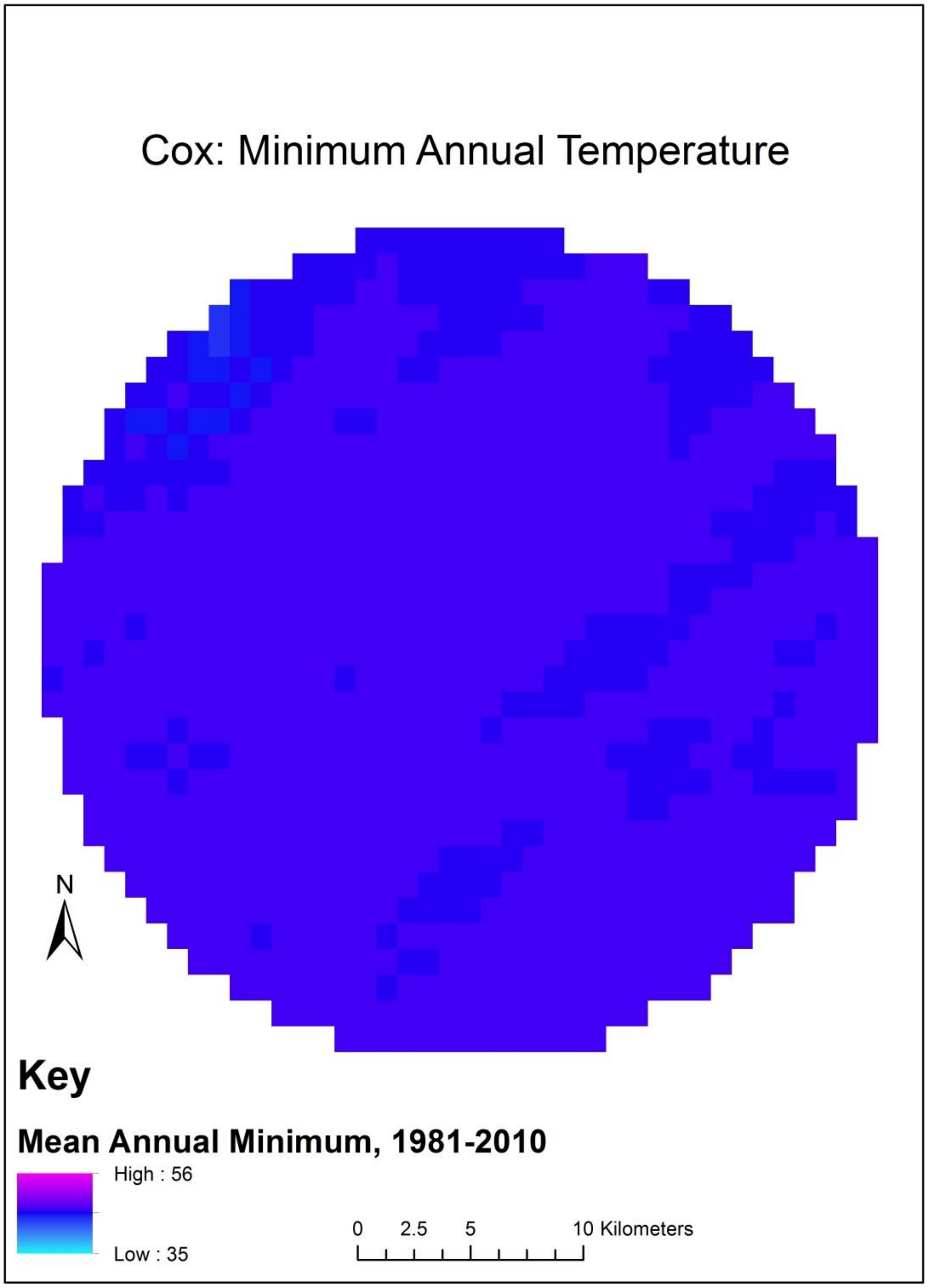

Figure 43. Temperature at Cox.

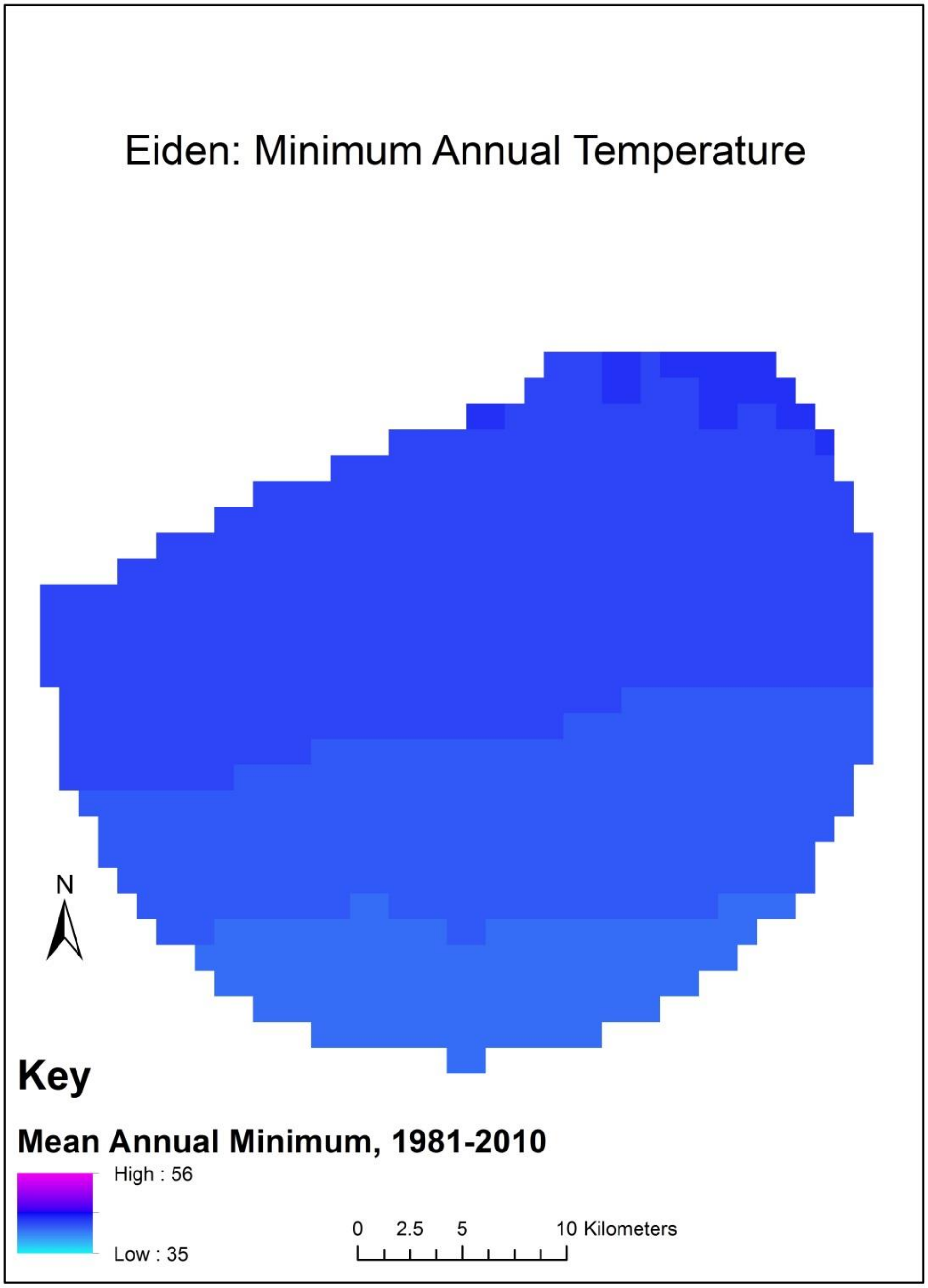

Figure 44. Temperature at Eiden.

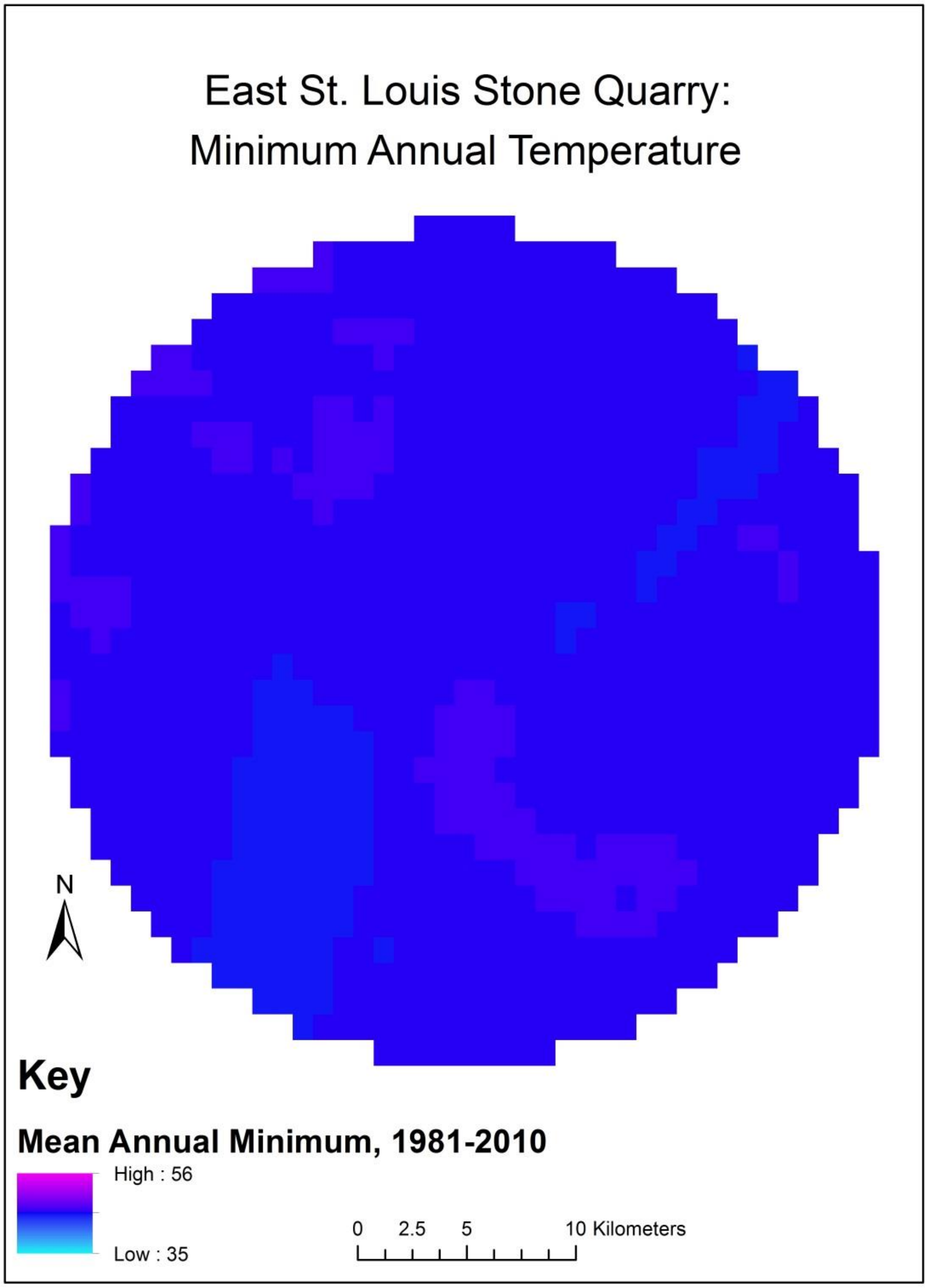

Figure 45. Temperature at East St. Louis Stone Quarry.

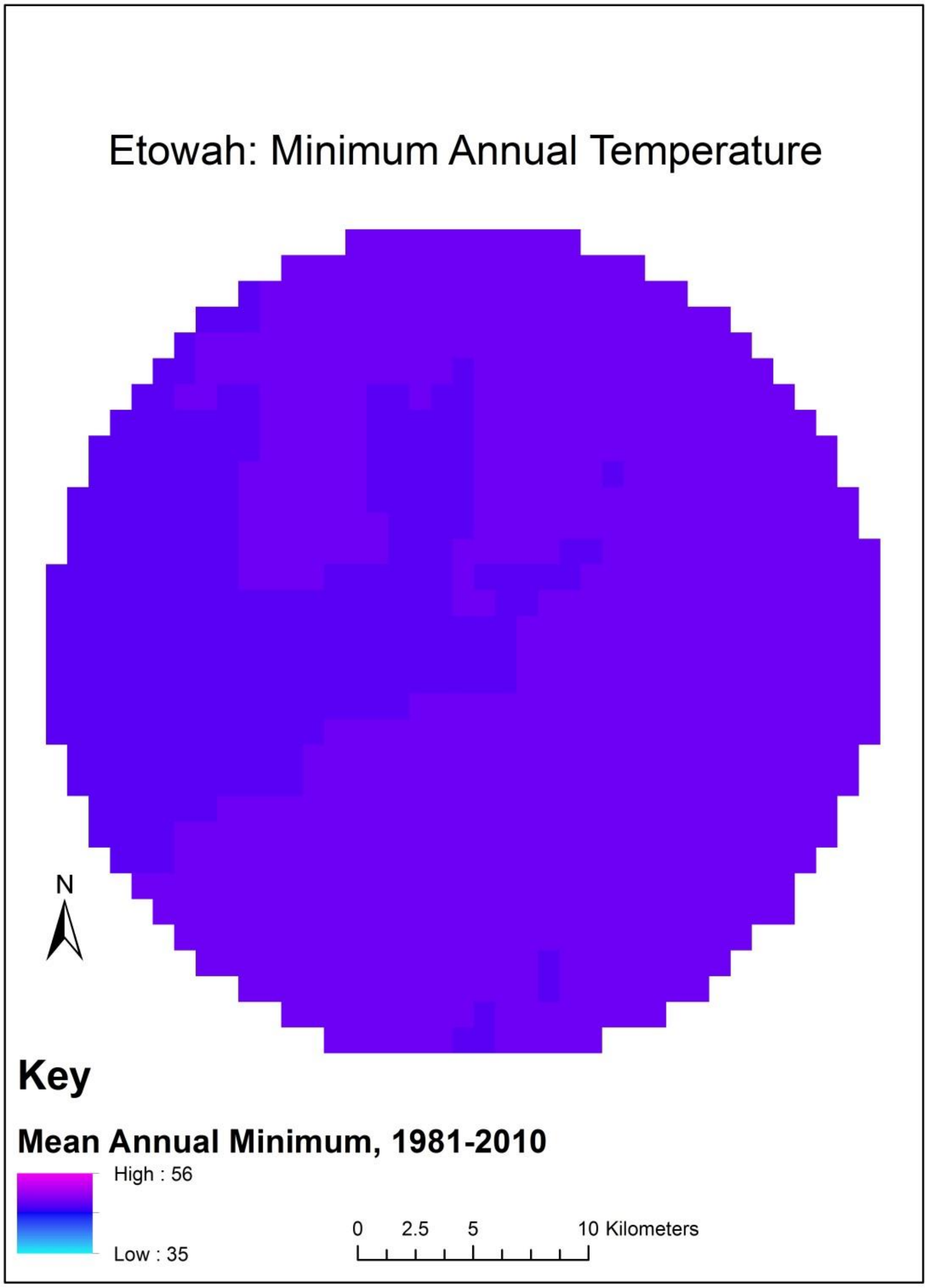

Figure 46. Temperature at Etowah.

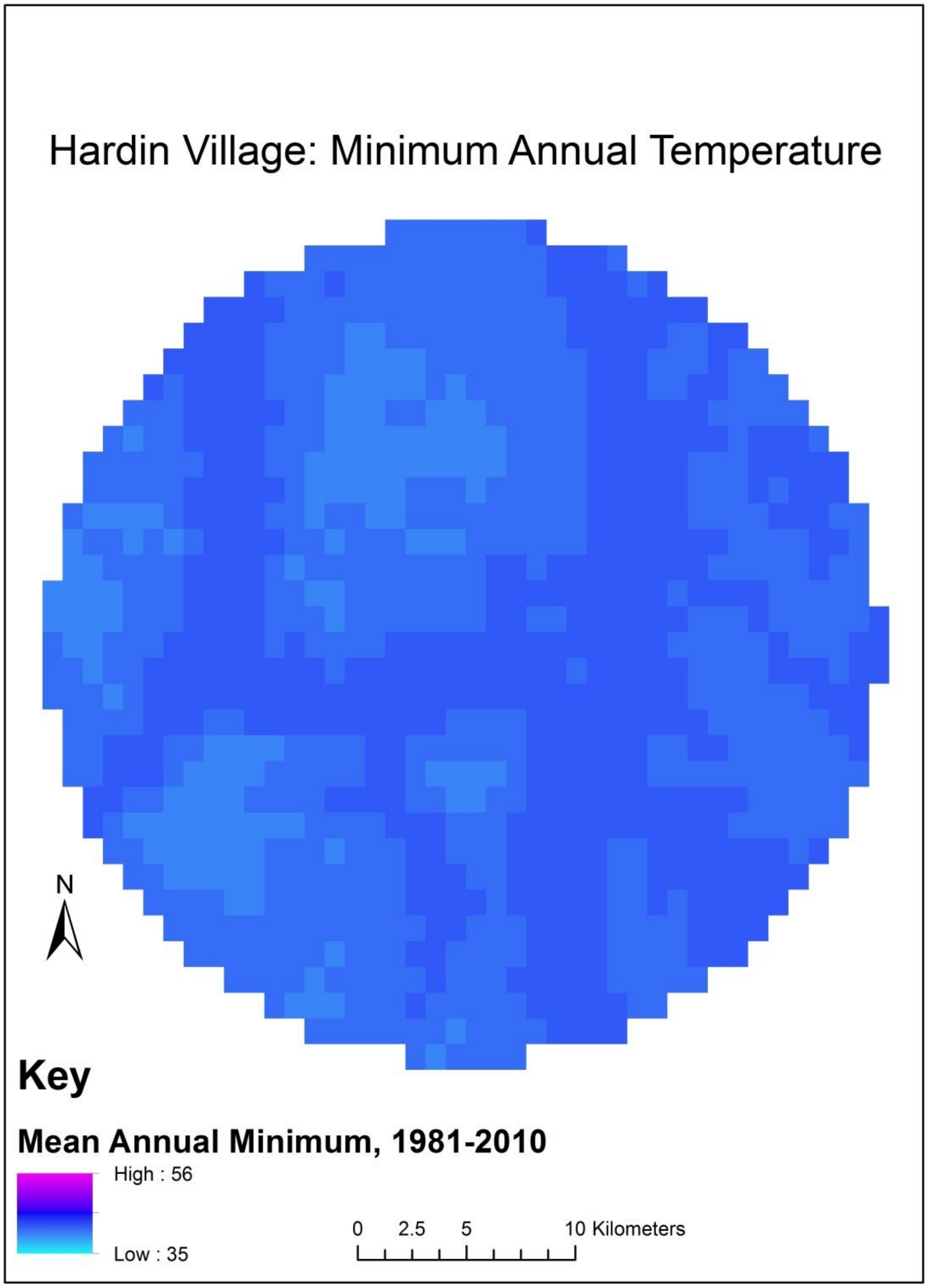

Figure 47. Temperature at Hardin Village.

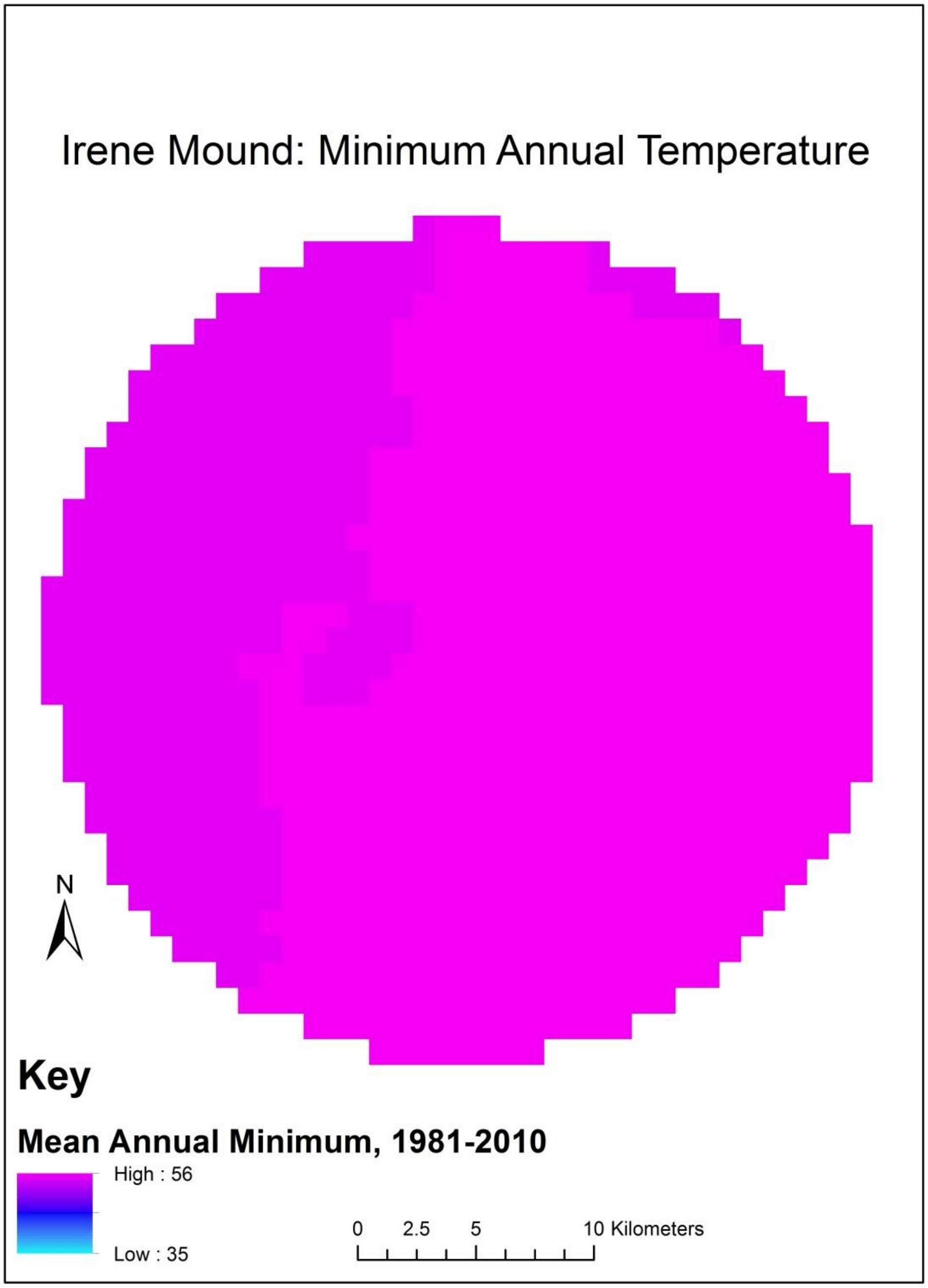

Figure 48. Temperature at Irene Mound.

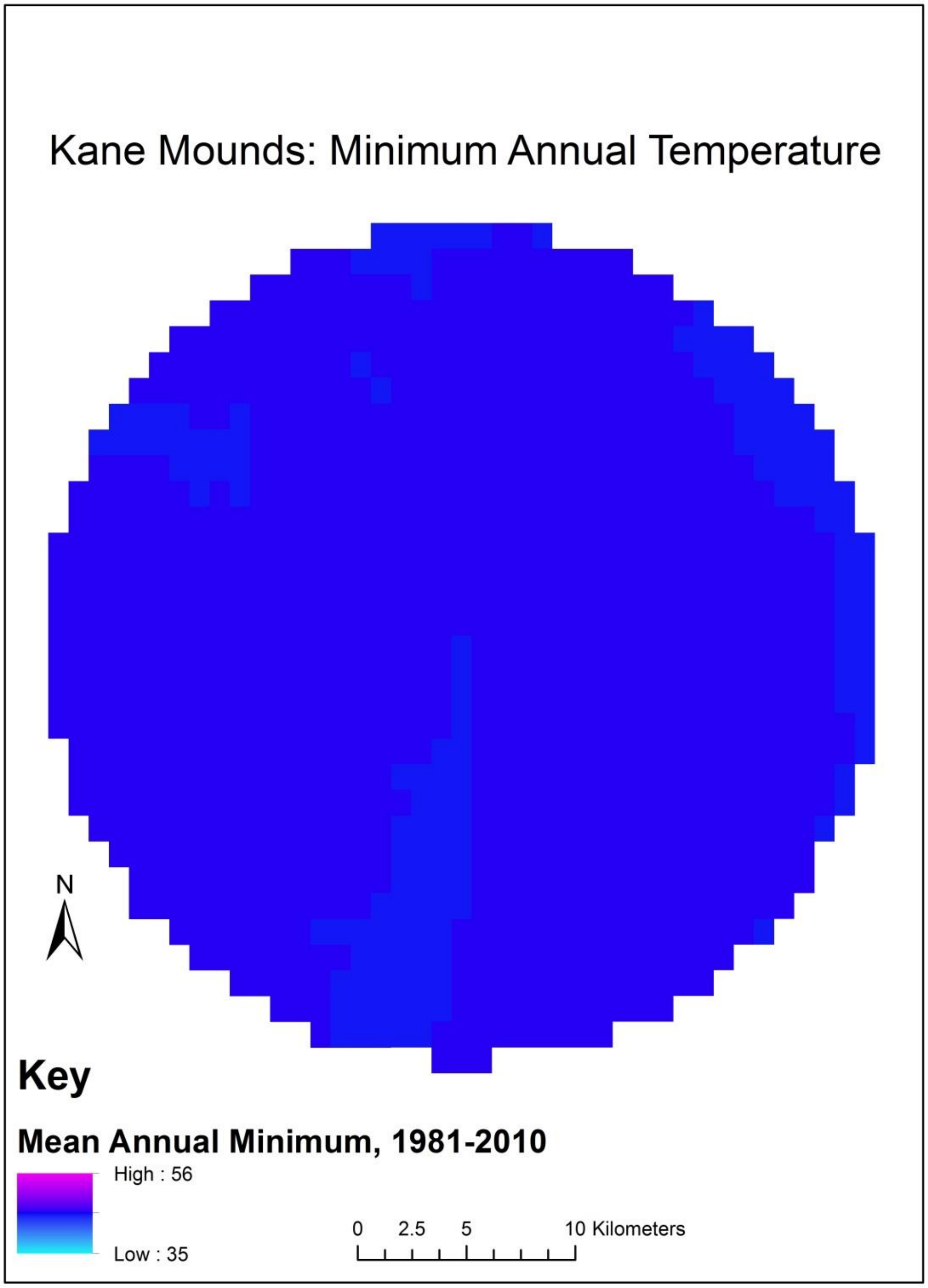

Figure 49. Temperature at Kane Mounds.

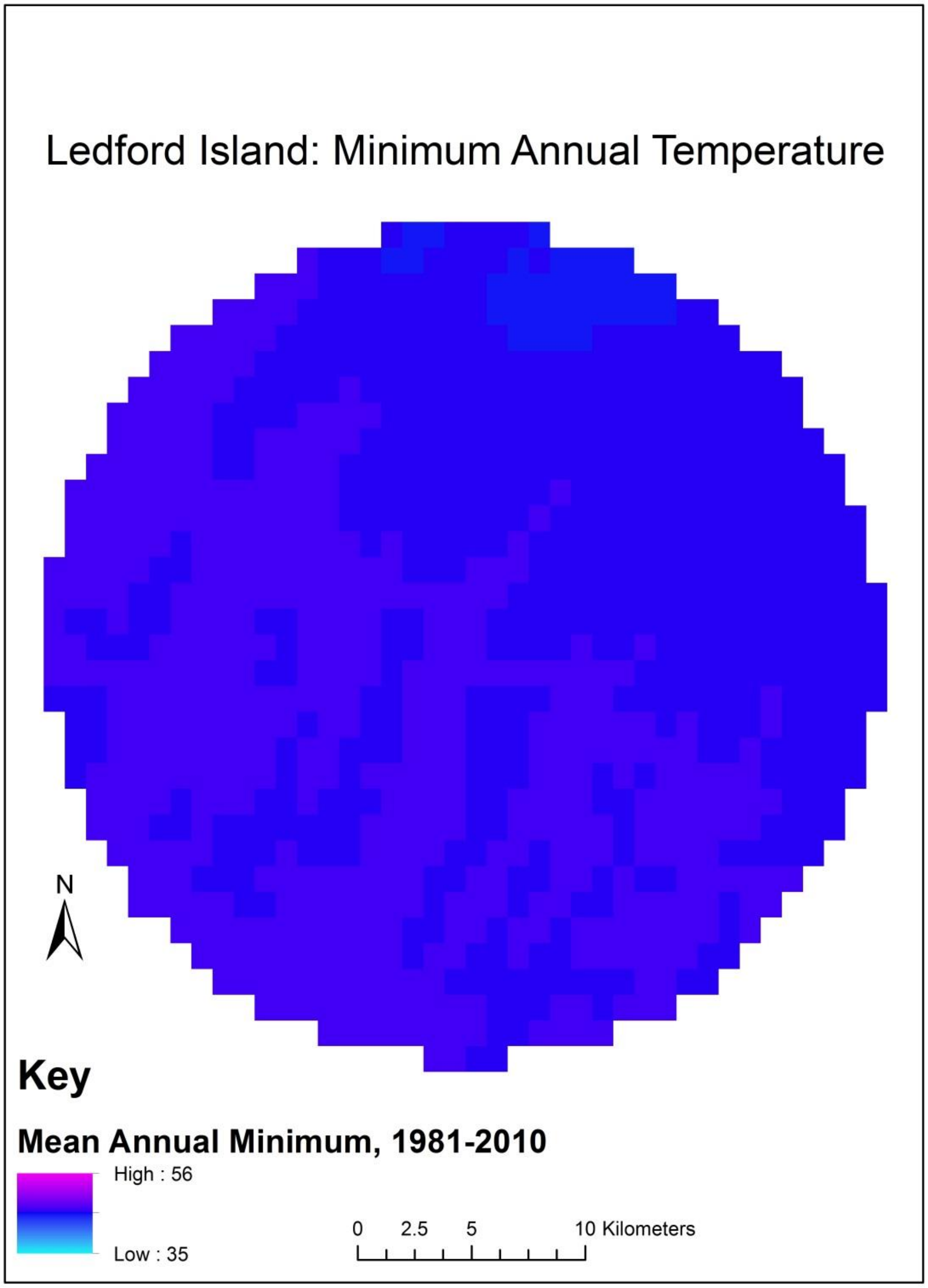

Figure 50. Temperature at Ledford Island.

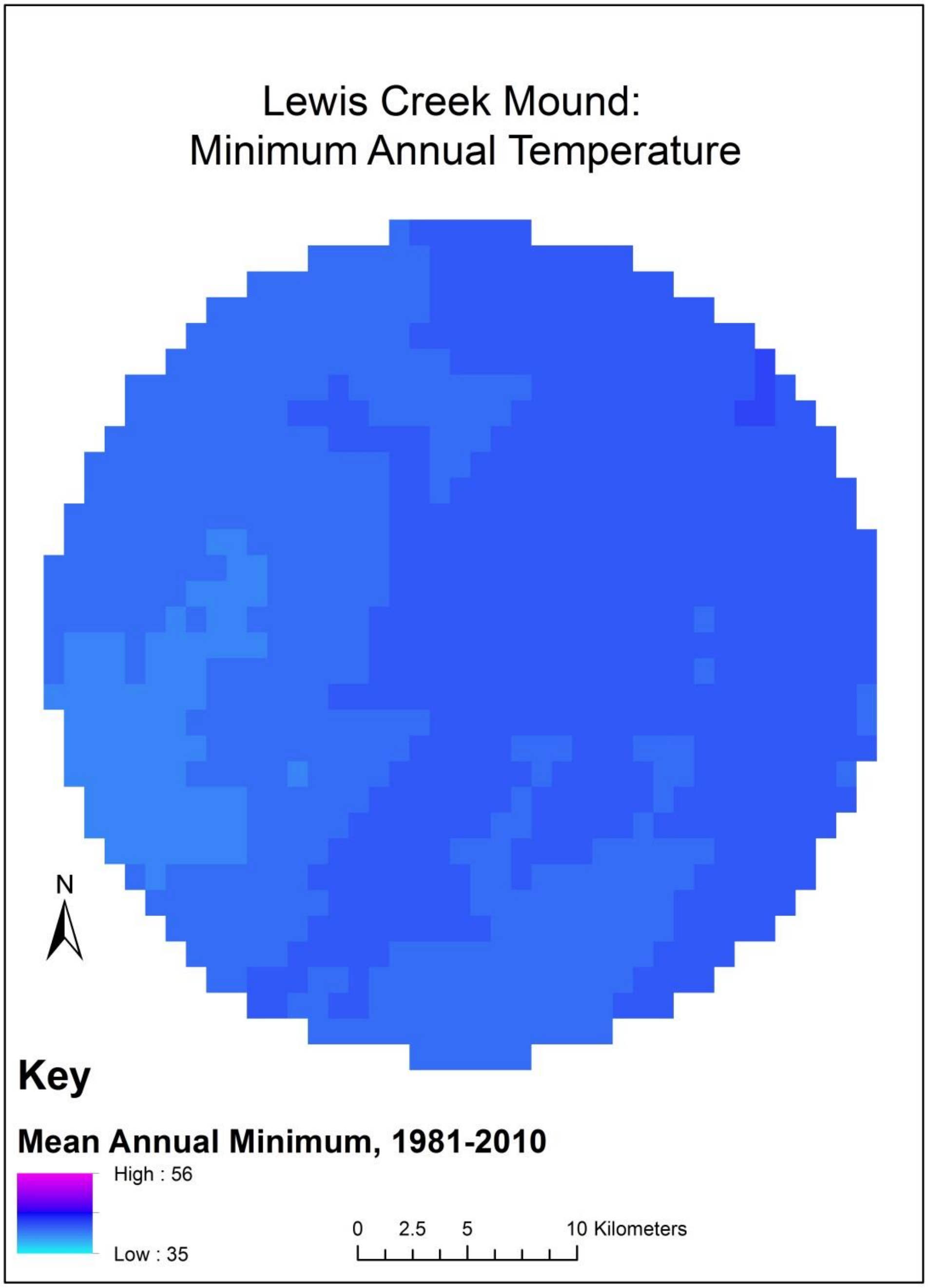

Figure 51. Temperature at Lewis Creek Mound.

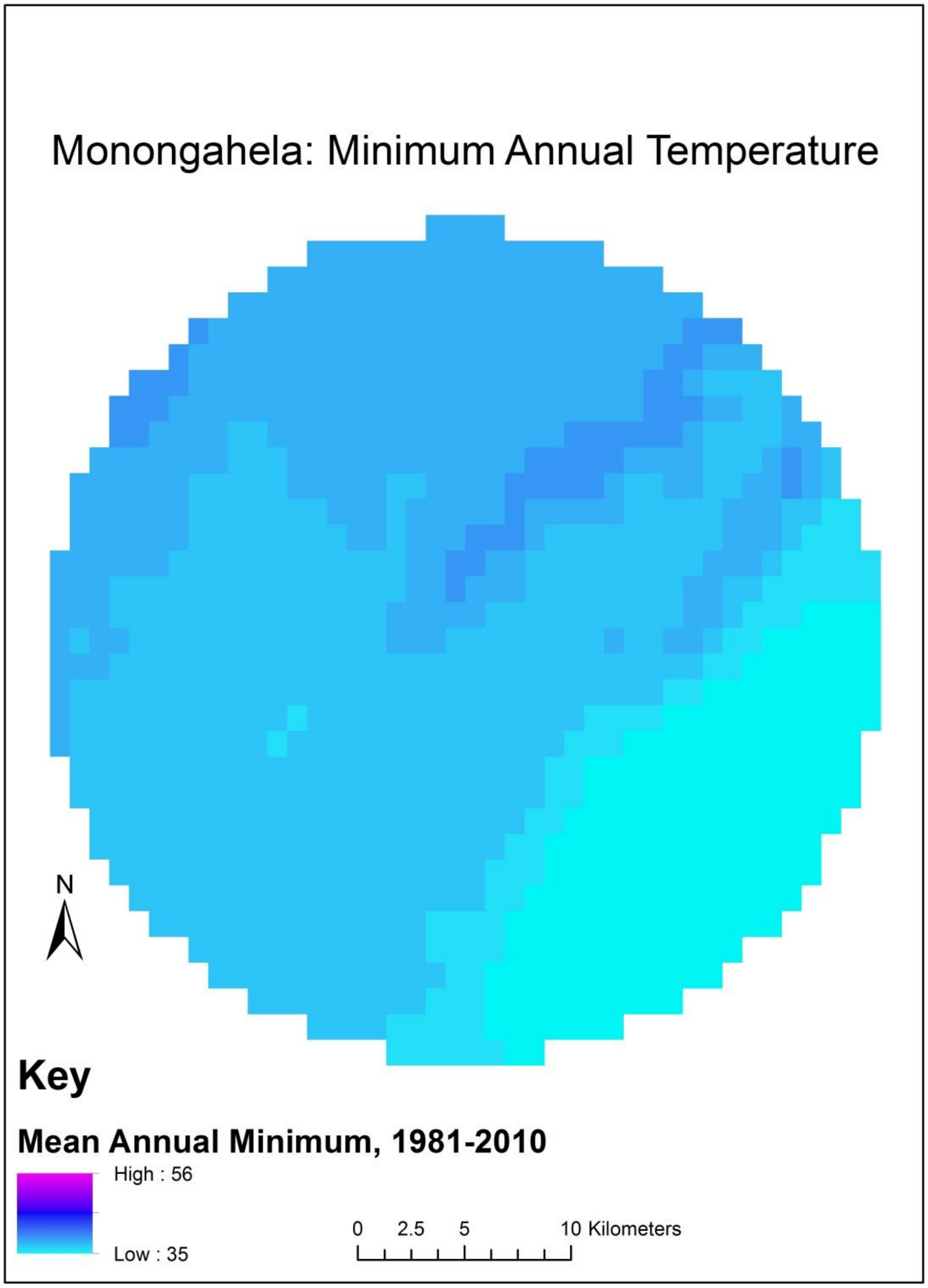

Figure 52. Temperature at Monongahela.

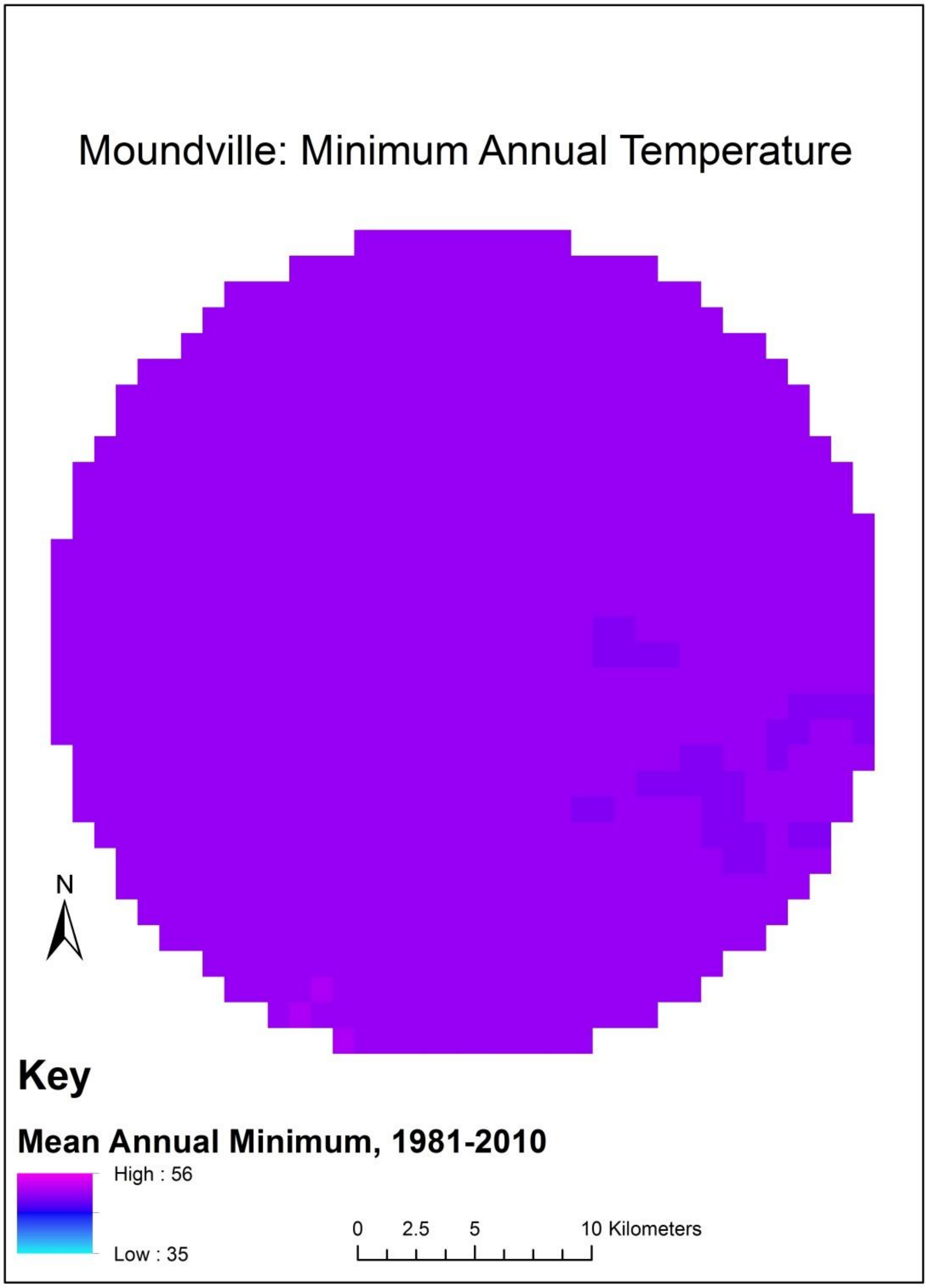

Figure 53. Temperature at Moundville.

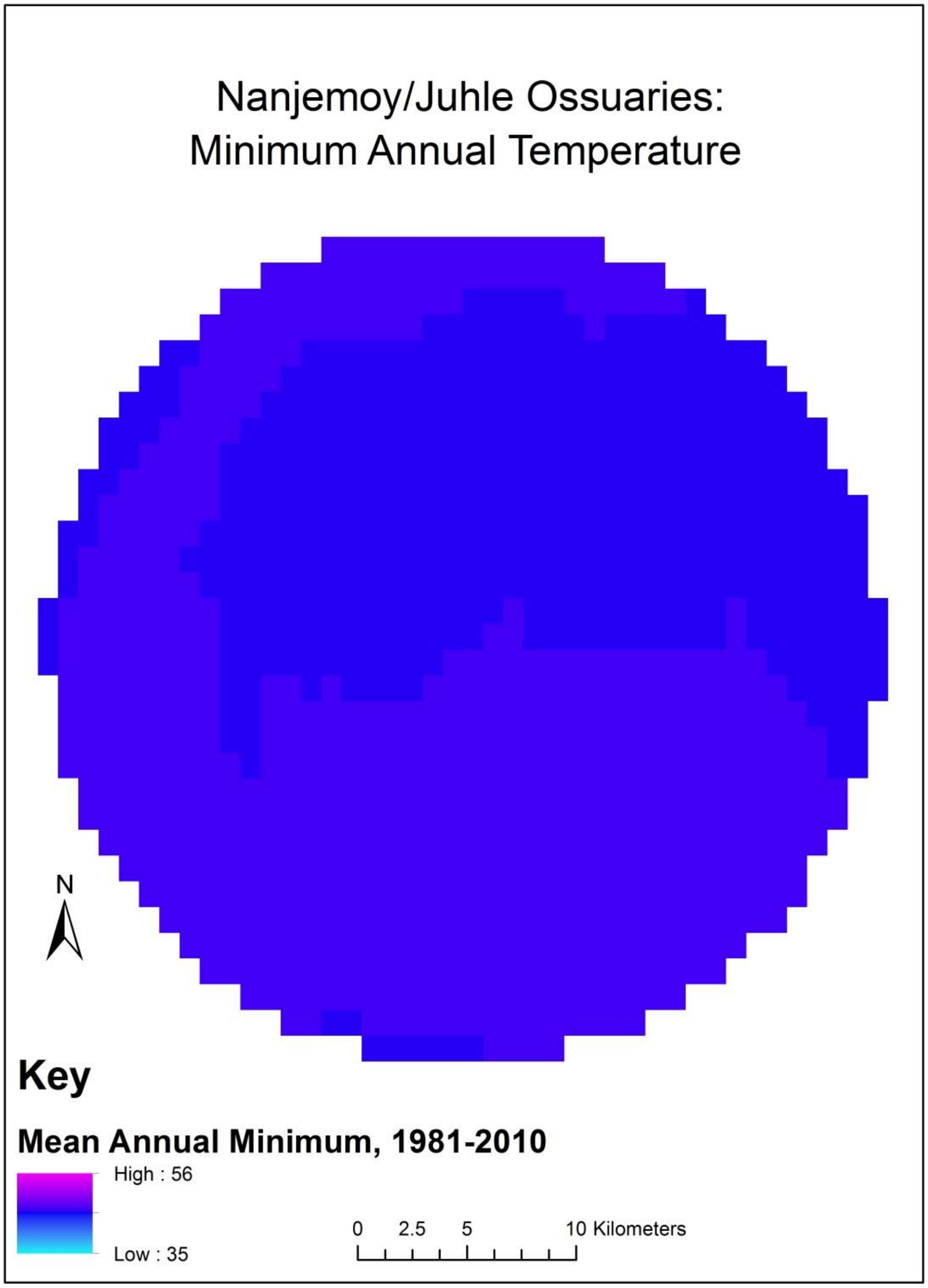

Figure 54. Temperature at Juhle Ossuary.

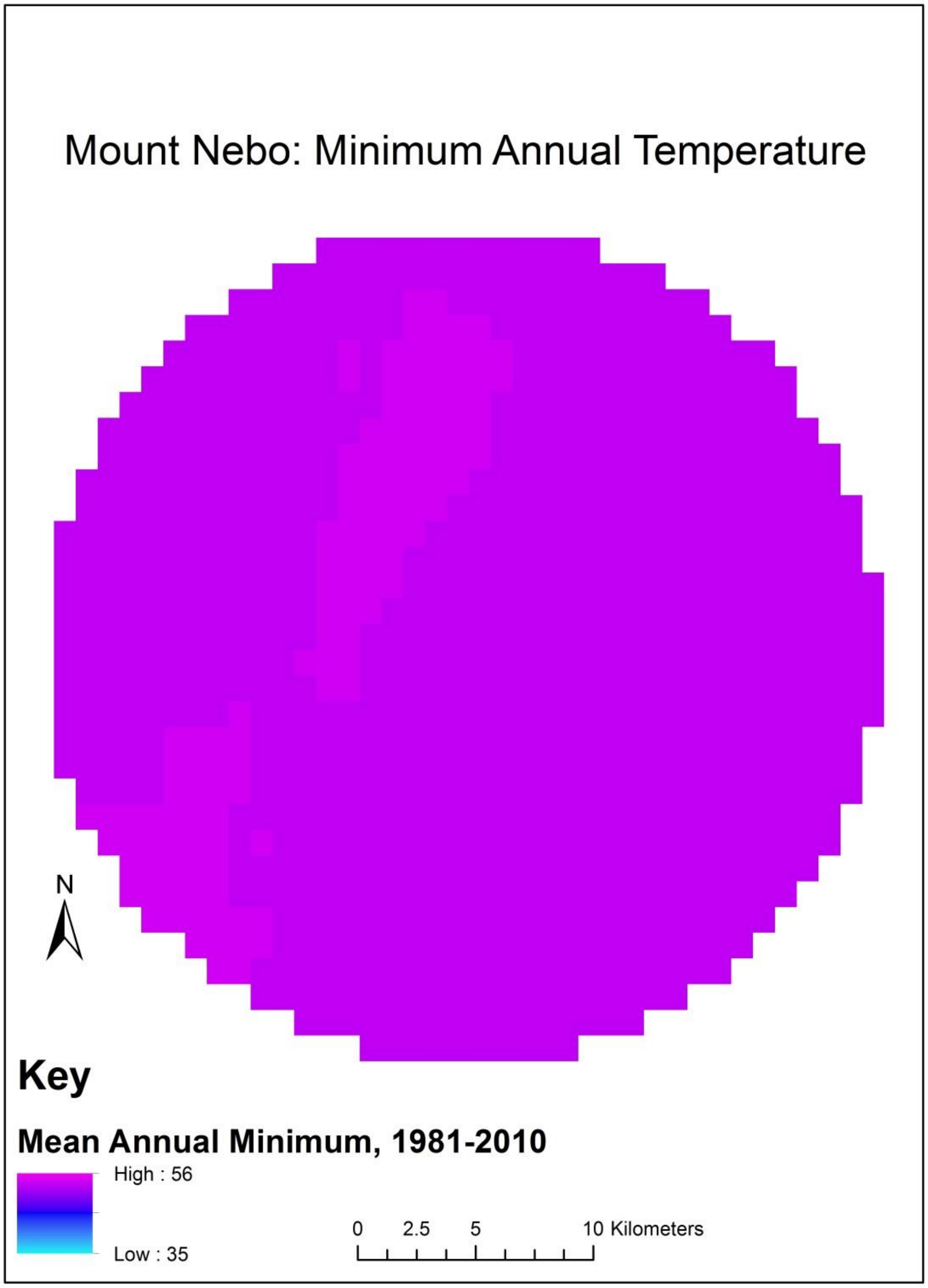

Figure 55. Temperature at Mount Nebo.

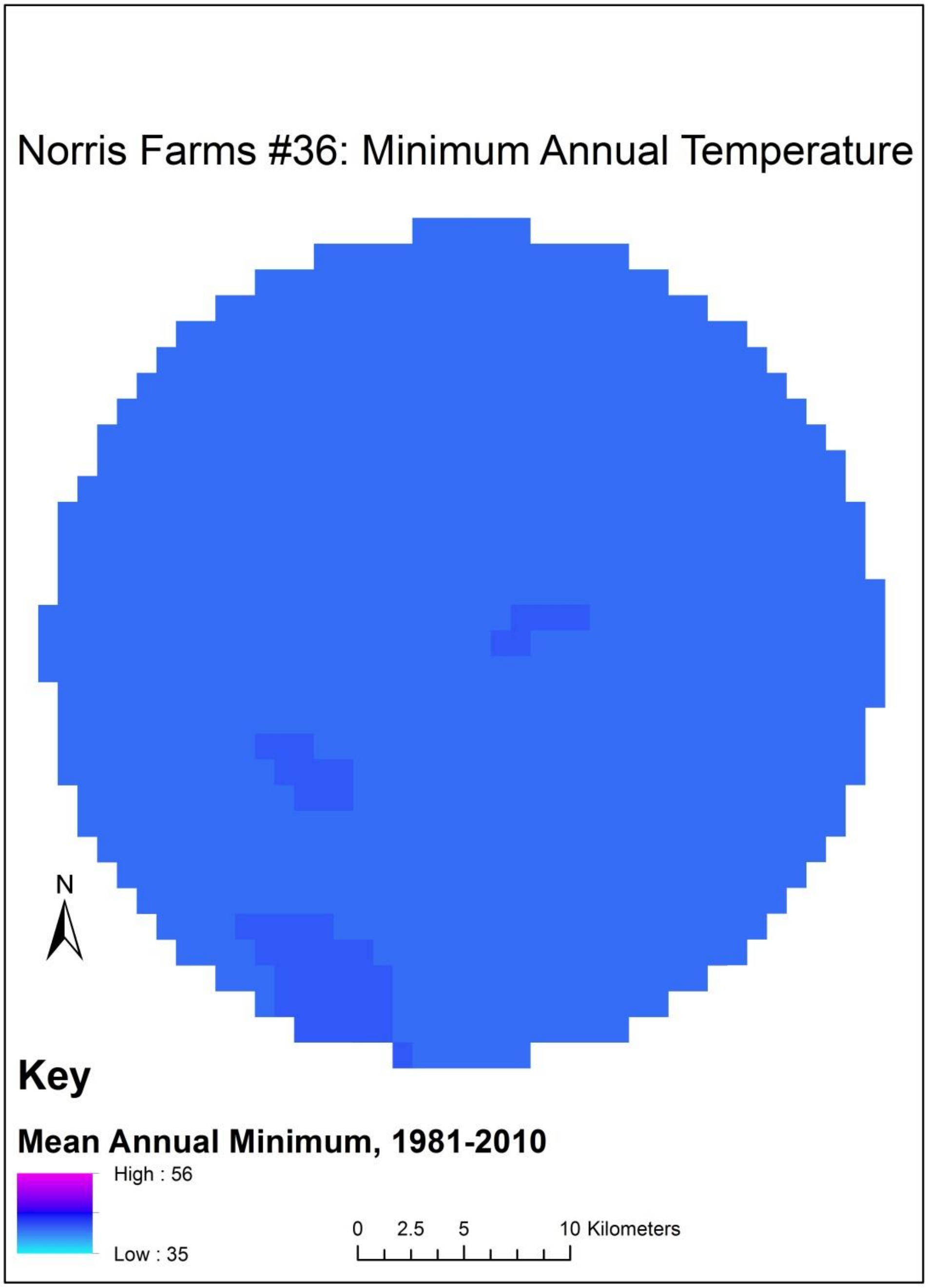

Figure 56. Temperature at Norris Farms #36.

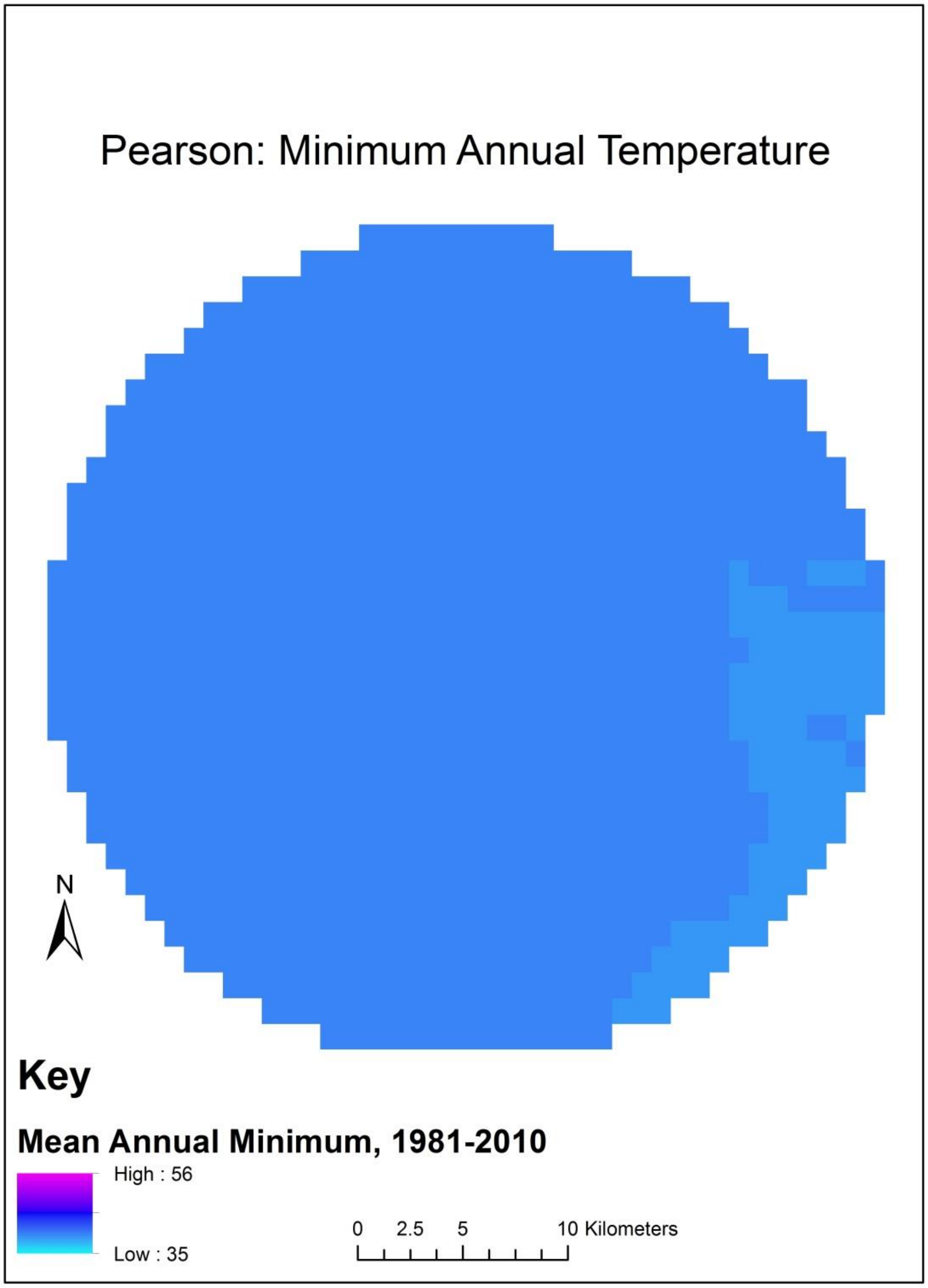

Figure 57. Temperature at Pearson.

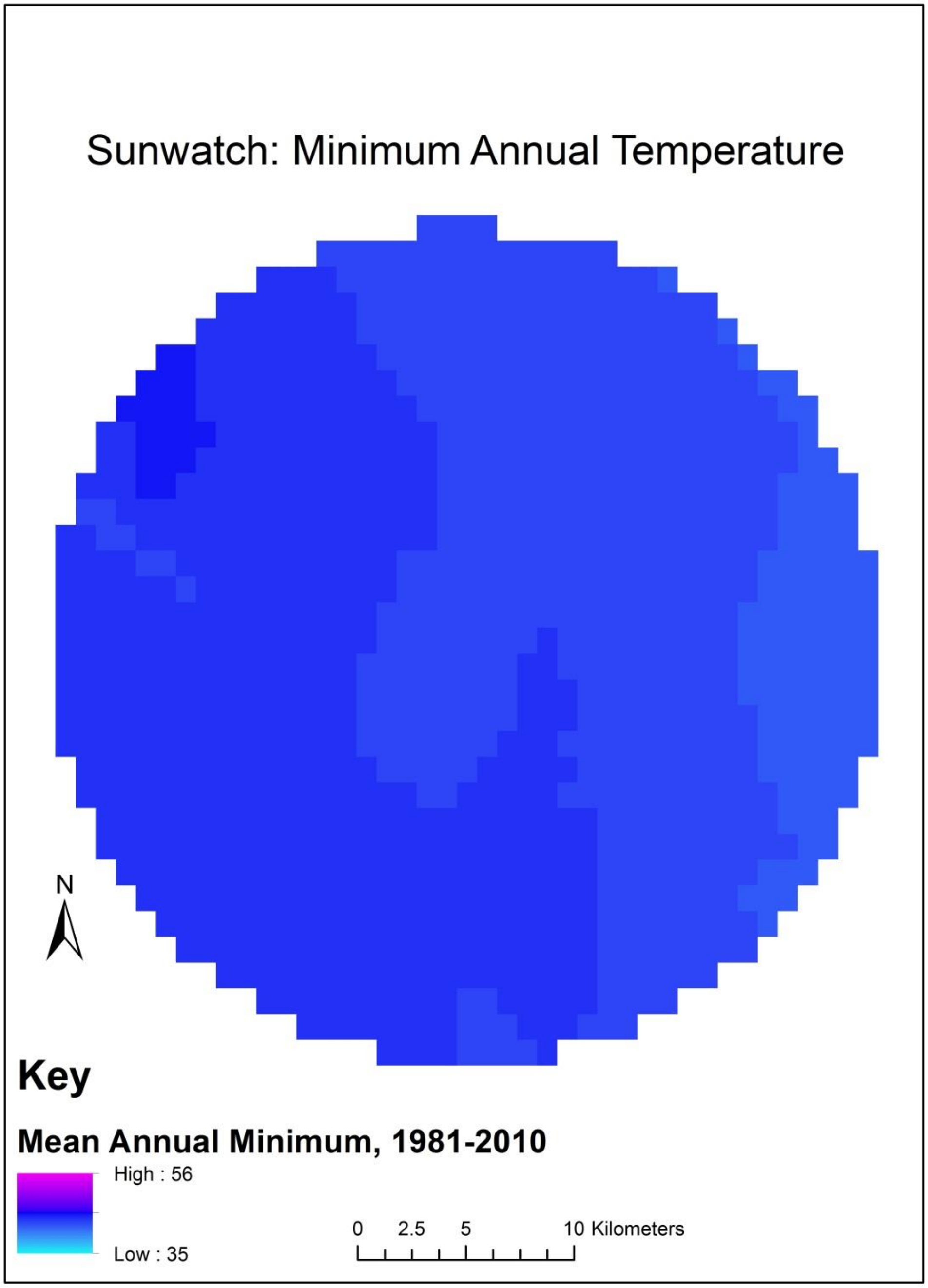

Figure 58. Temperature at Sunwatch.

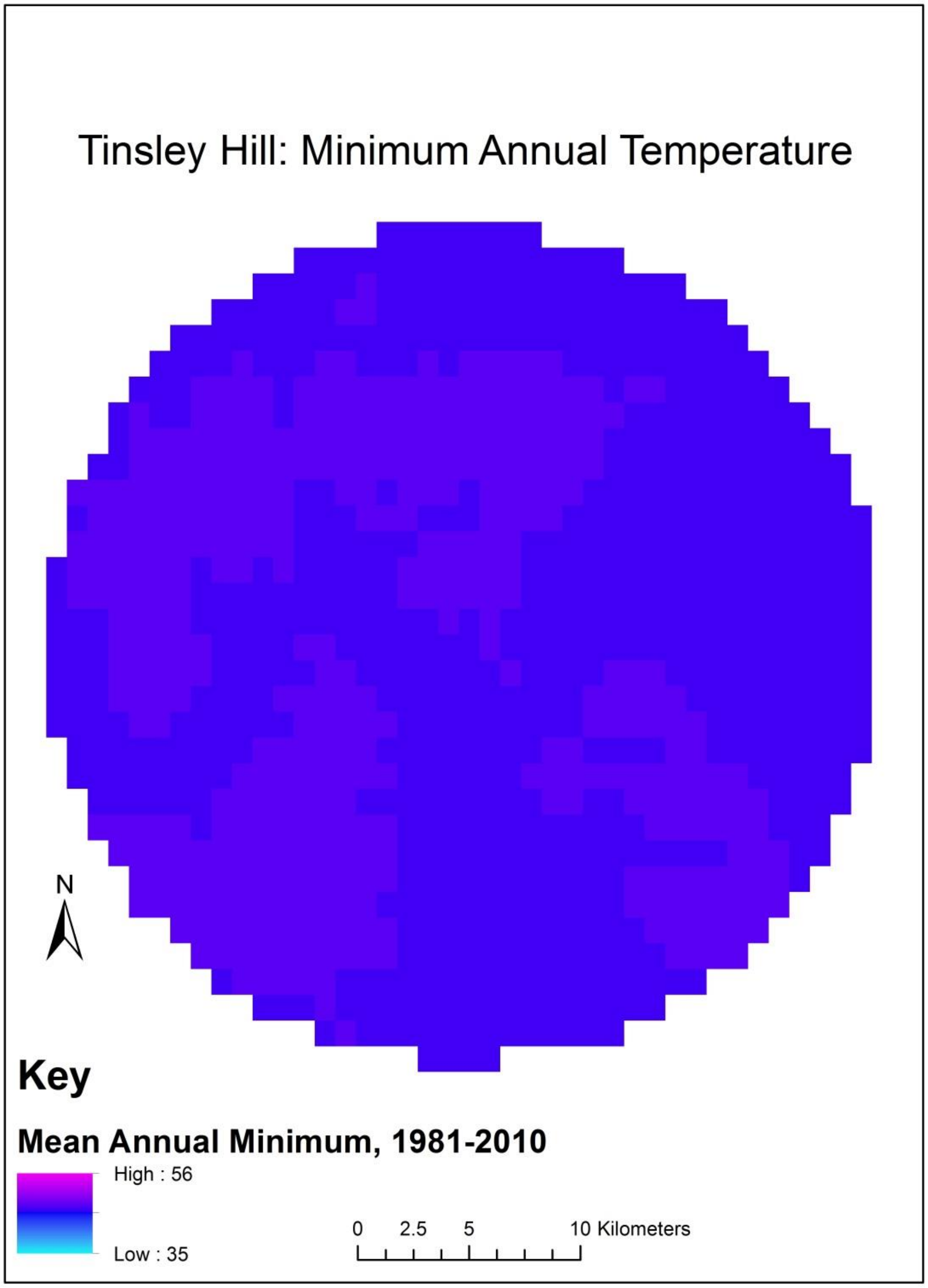

Figure 59. Temperature at Tinsley Hill.

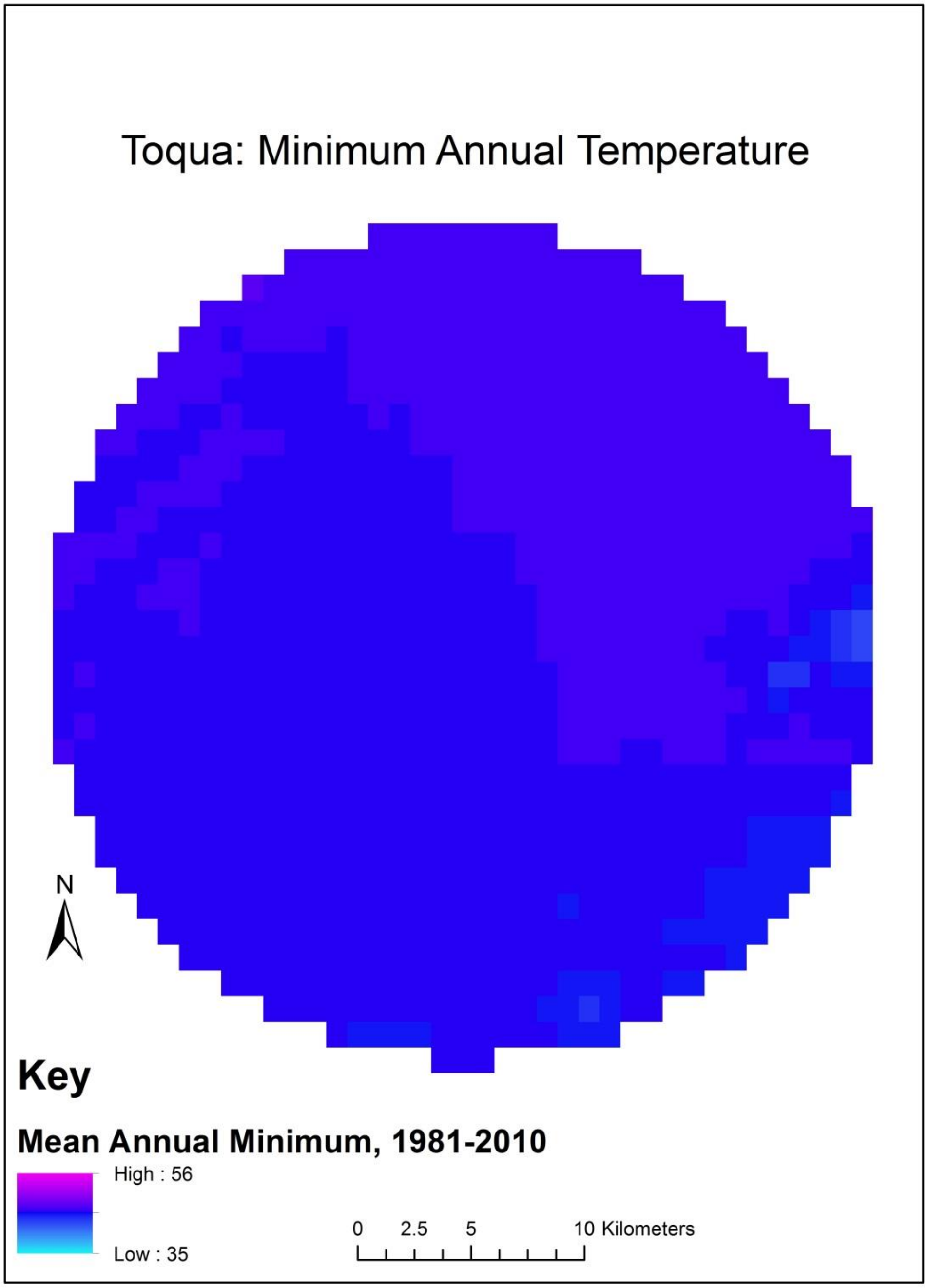

Figure 60. Temperature at Toqua.

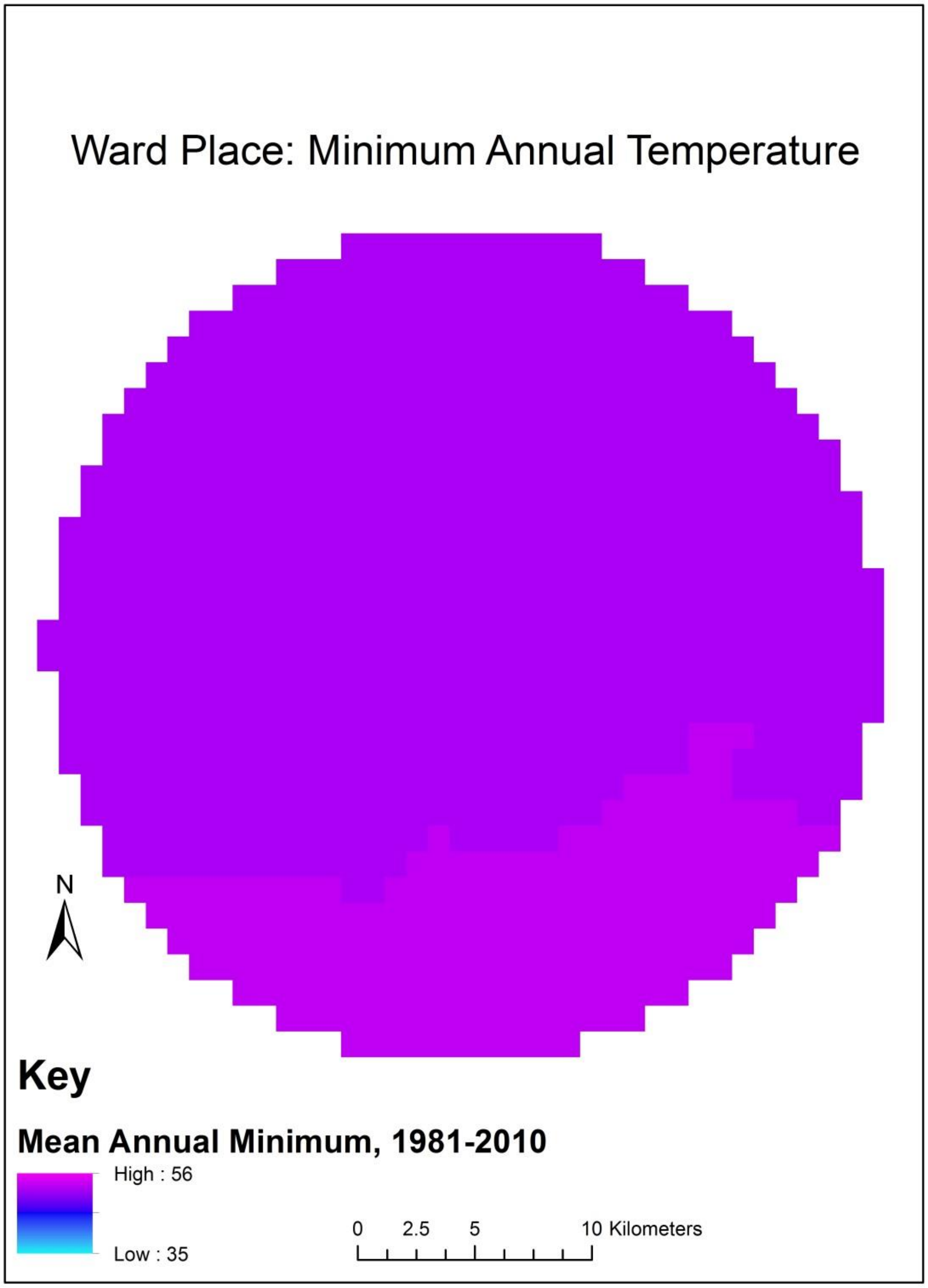

Figure 61. Temperature at Ward Place.

A3. Elevation

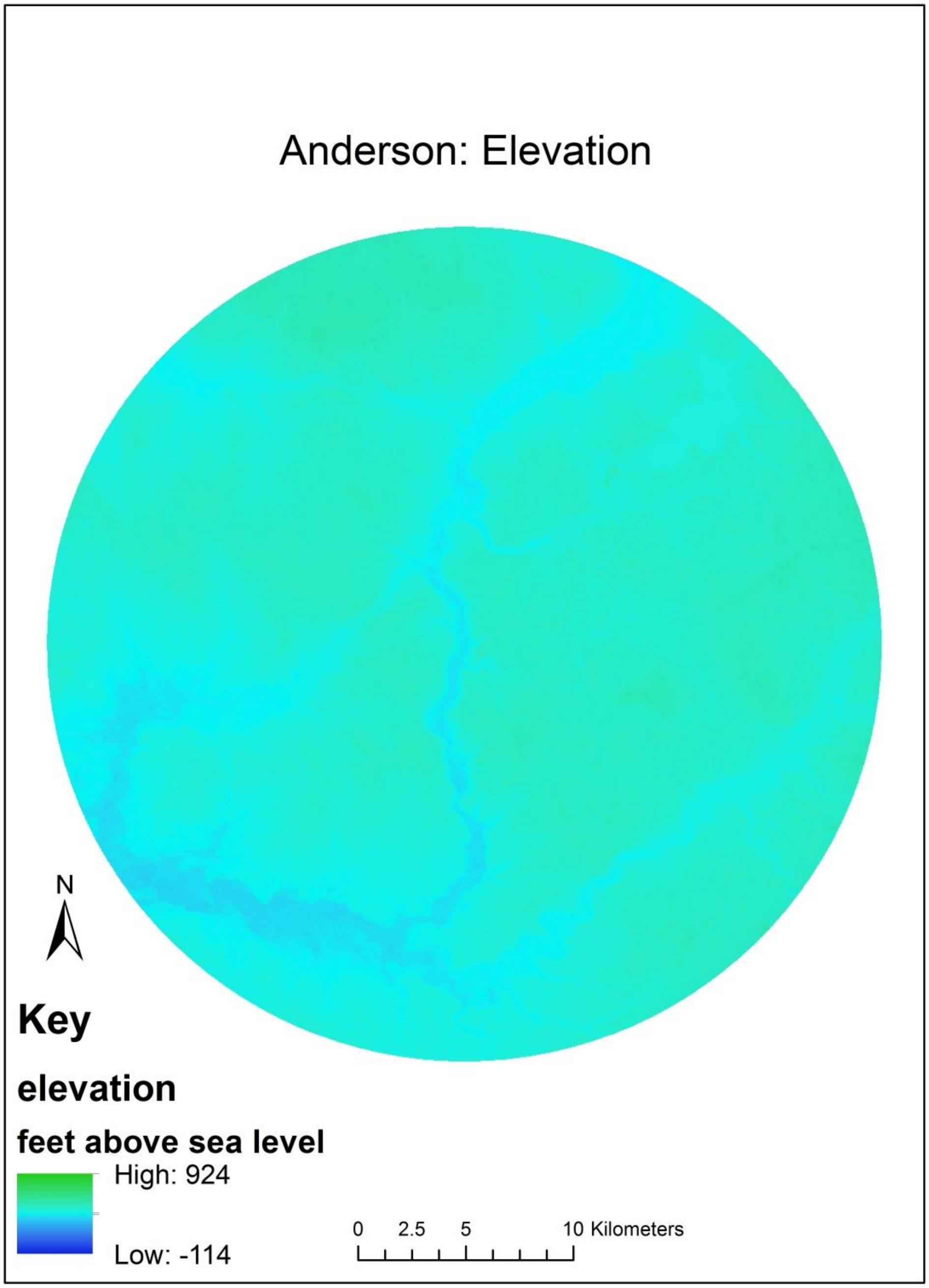

Figure 62. Elevation at Anderson.

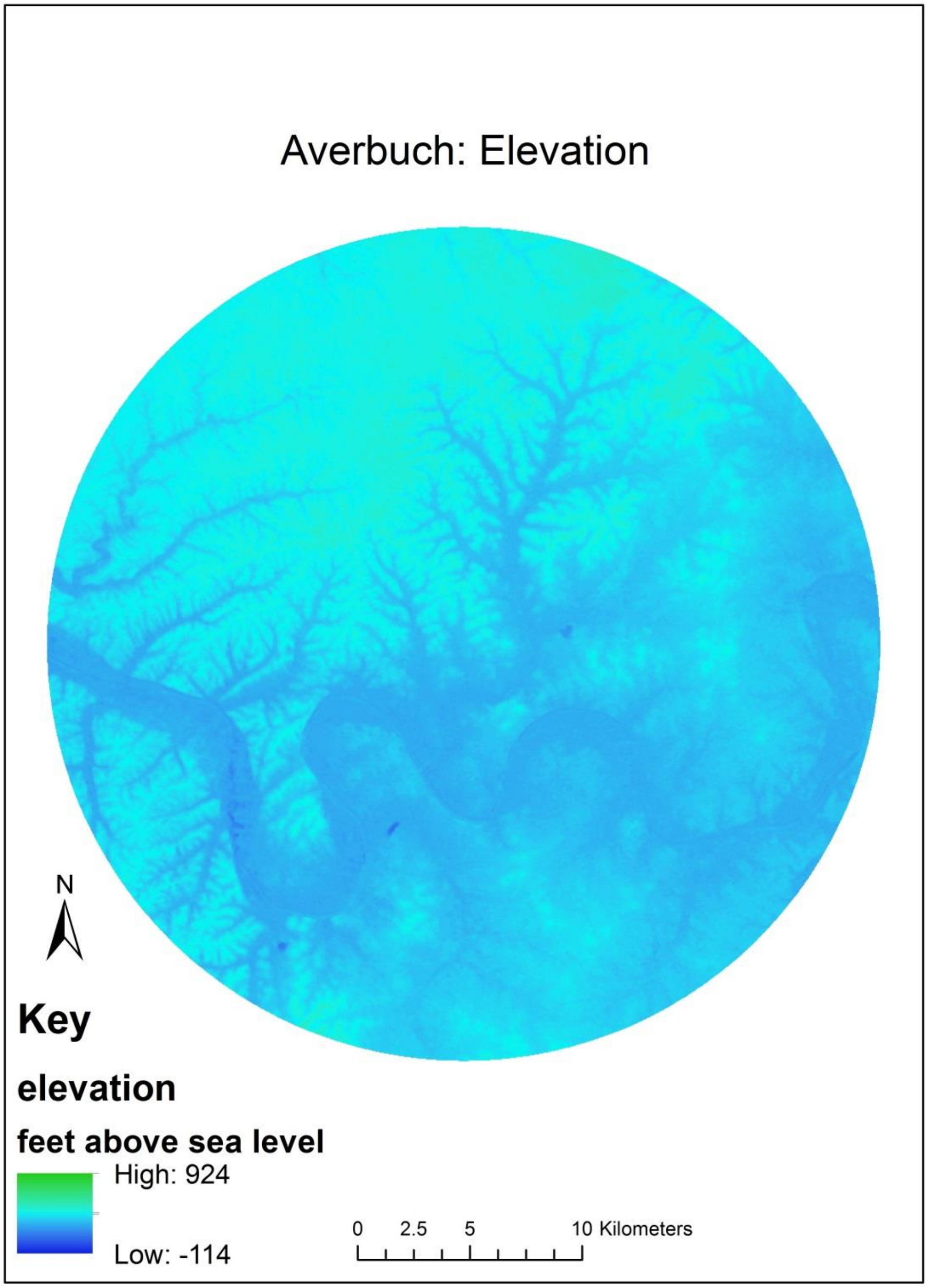

Figure 63. Elevation at Averbuch.

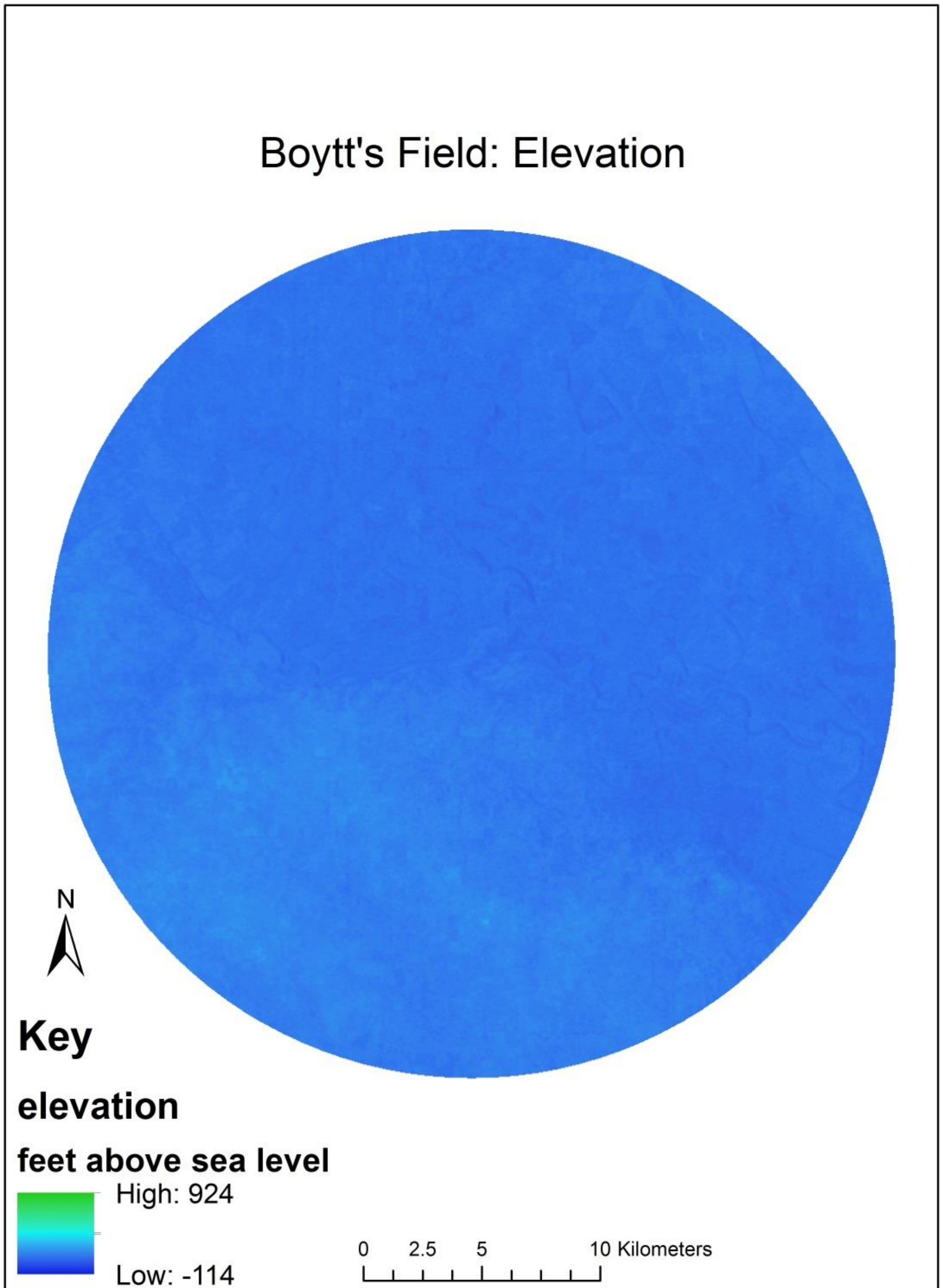

Figure 64. Elevation at Boytt's Field.

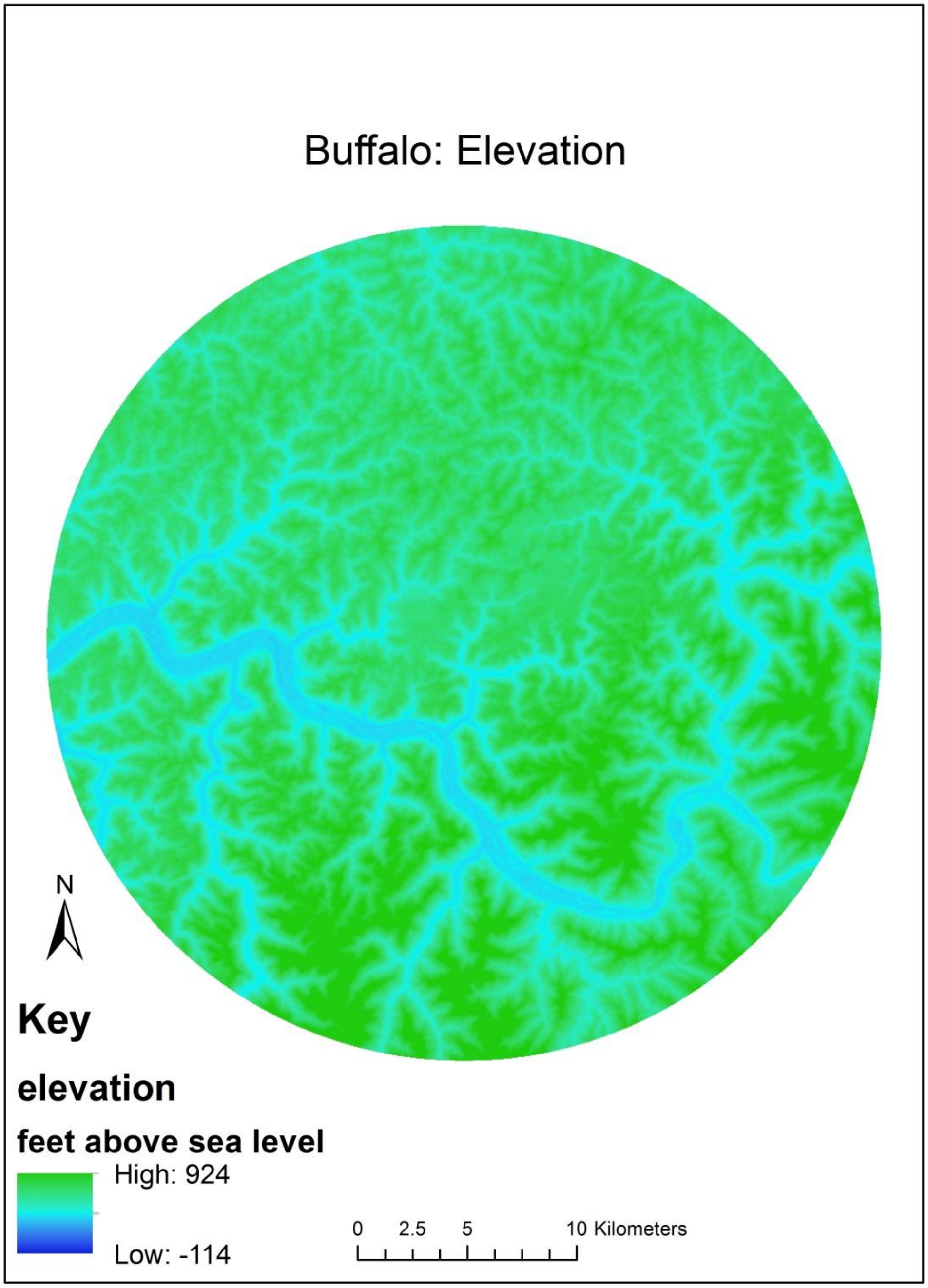

Figure 65. Elevation at Buffalo.

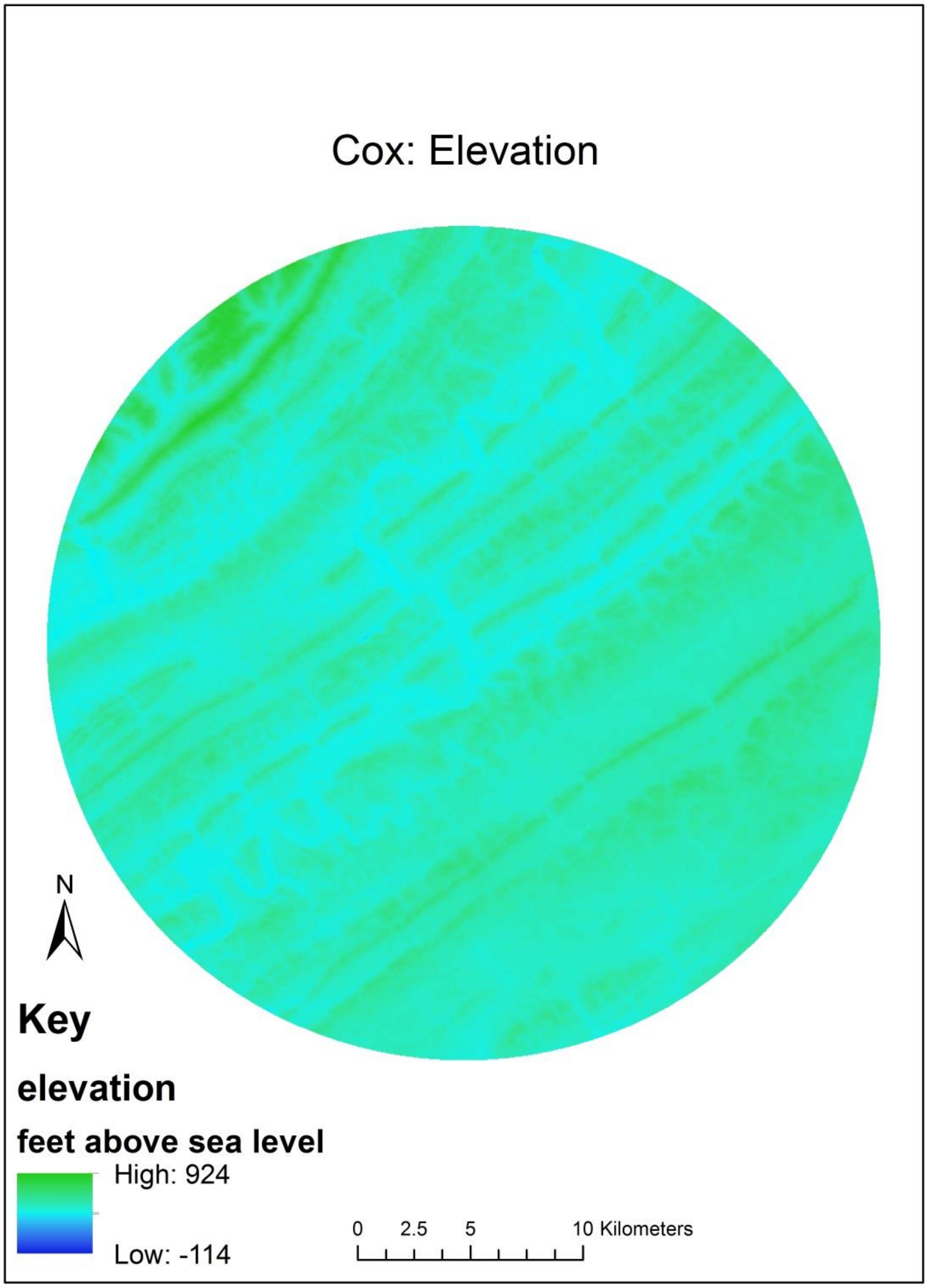

Figure 66. Elevation at Cox.

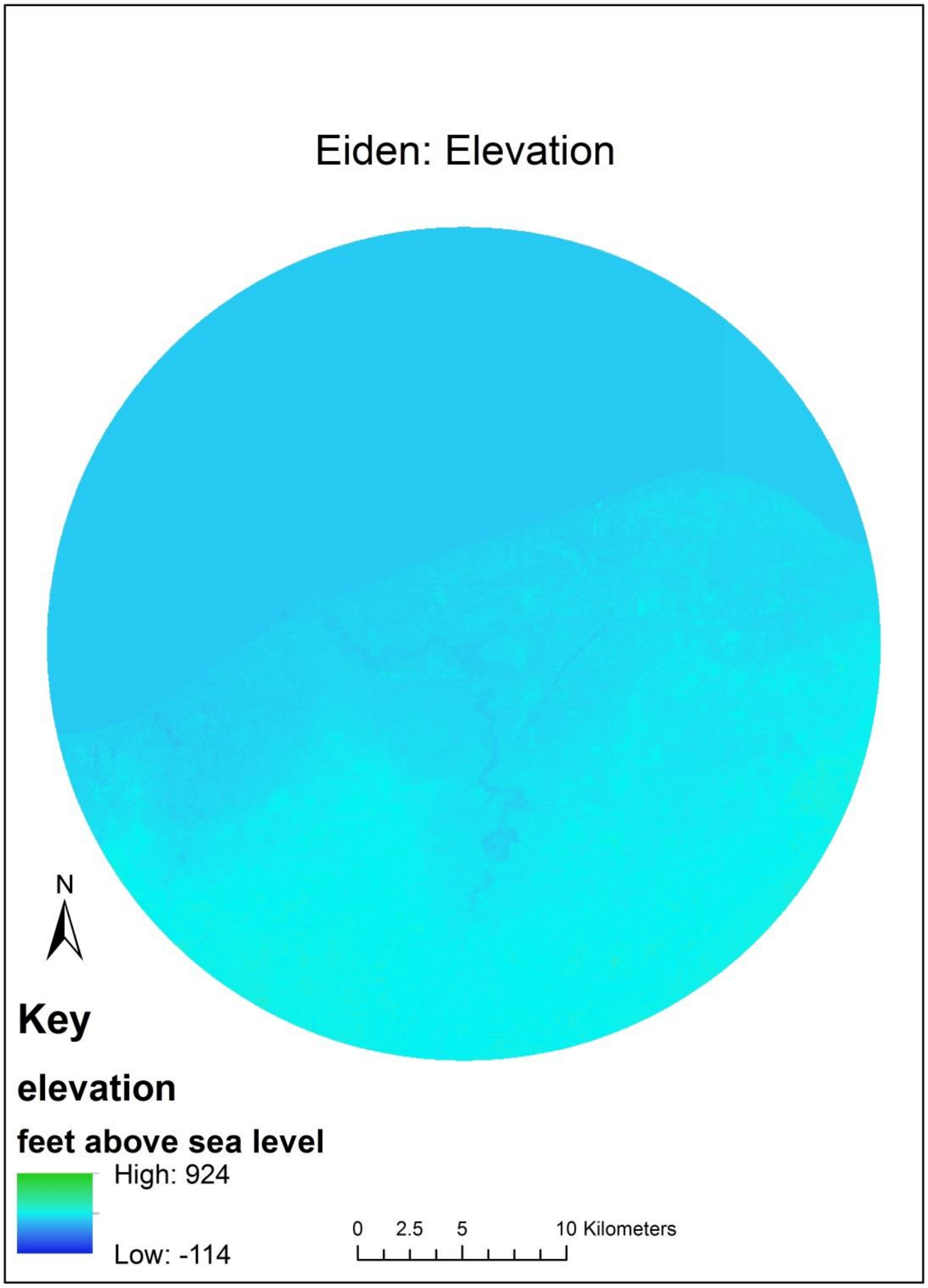

Figure 67. Elevation at Eiden.

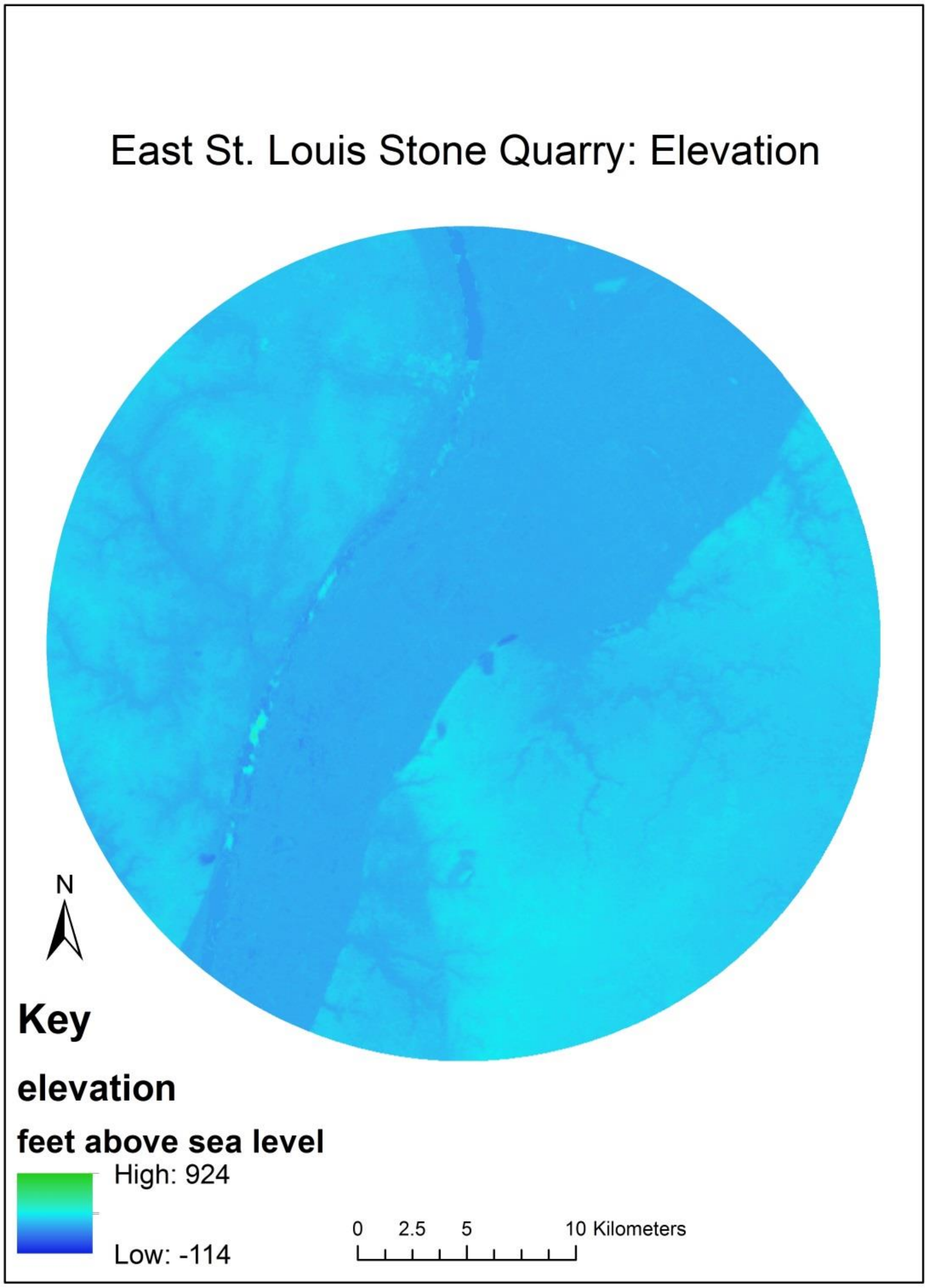

Figure 68. Elevation at East St. Louis Stone Quarry.

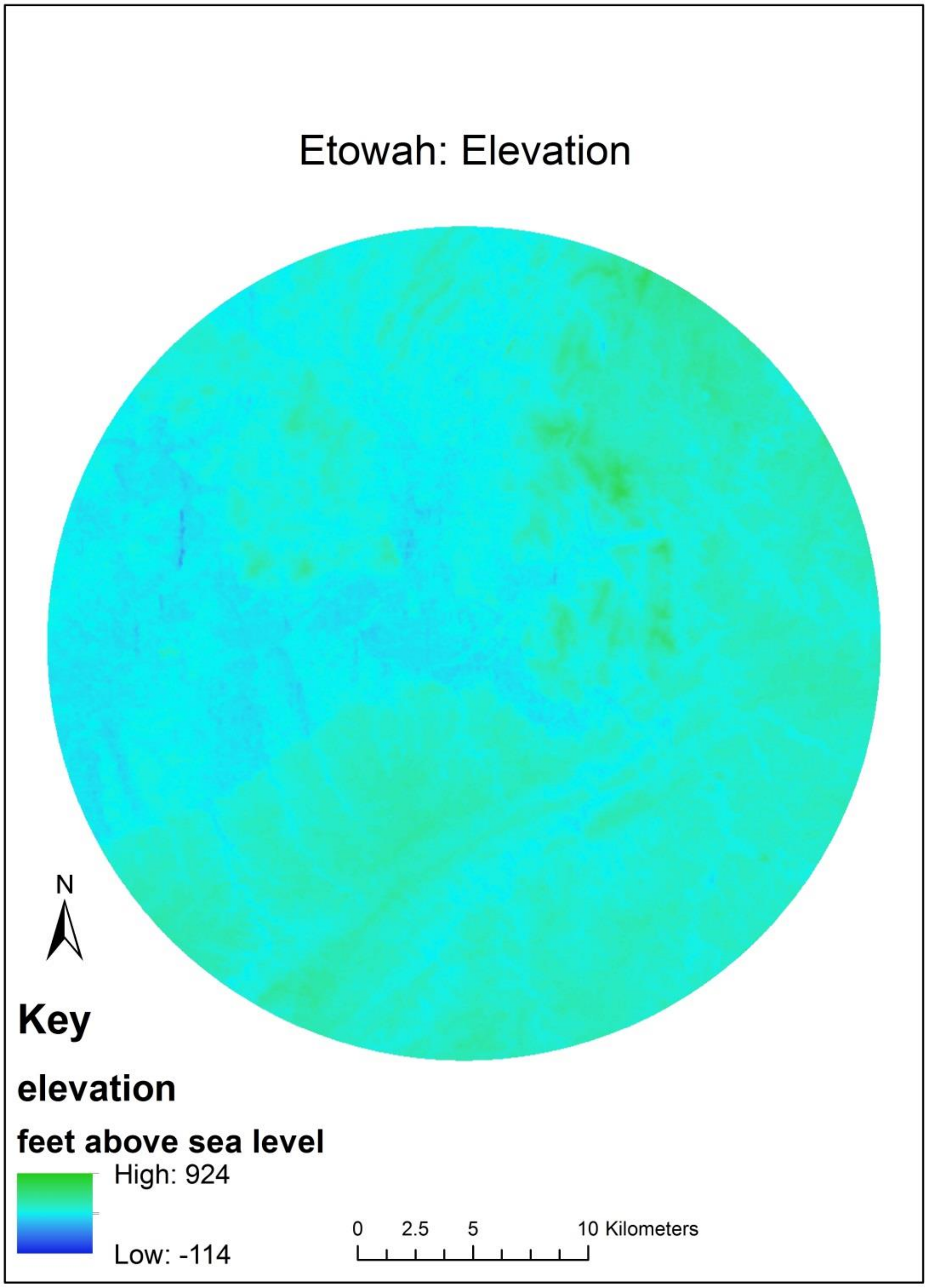

Figure 69. Elevation at Etowah.

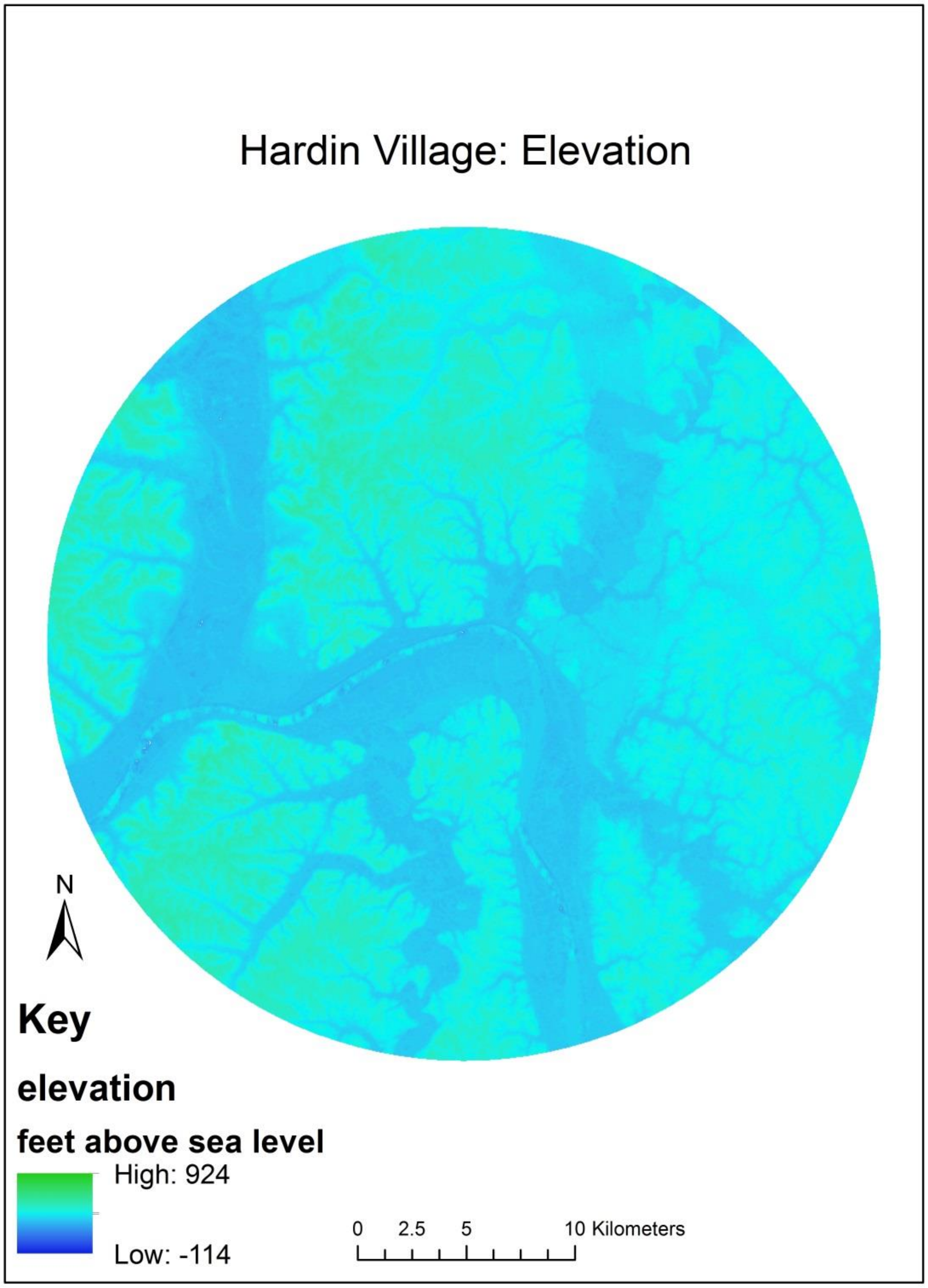

Figure 70. Elevation at Hardin Village.

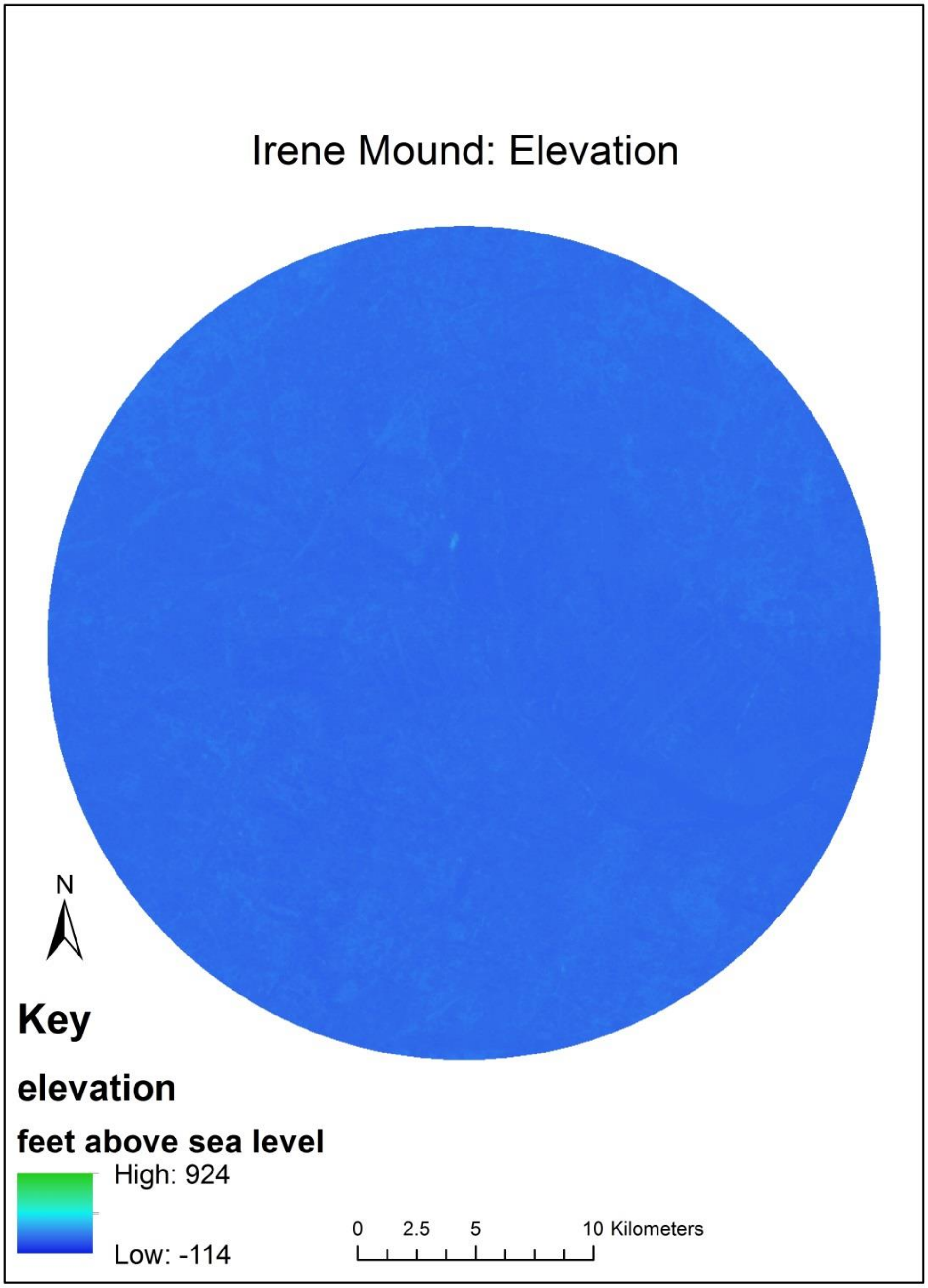

Figure 71. Elevation at Irene Mound.

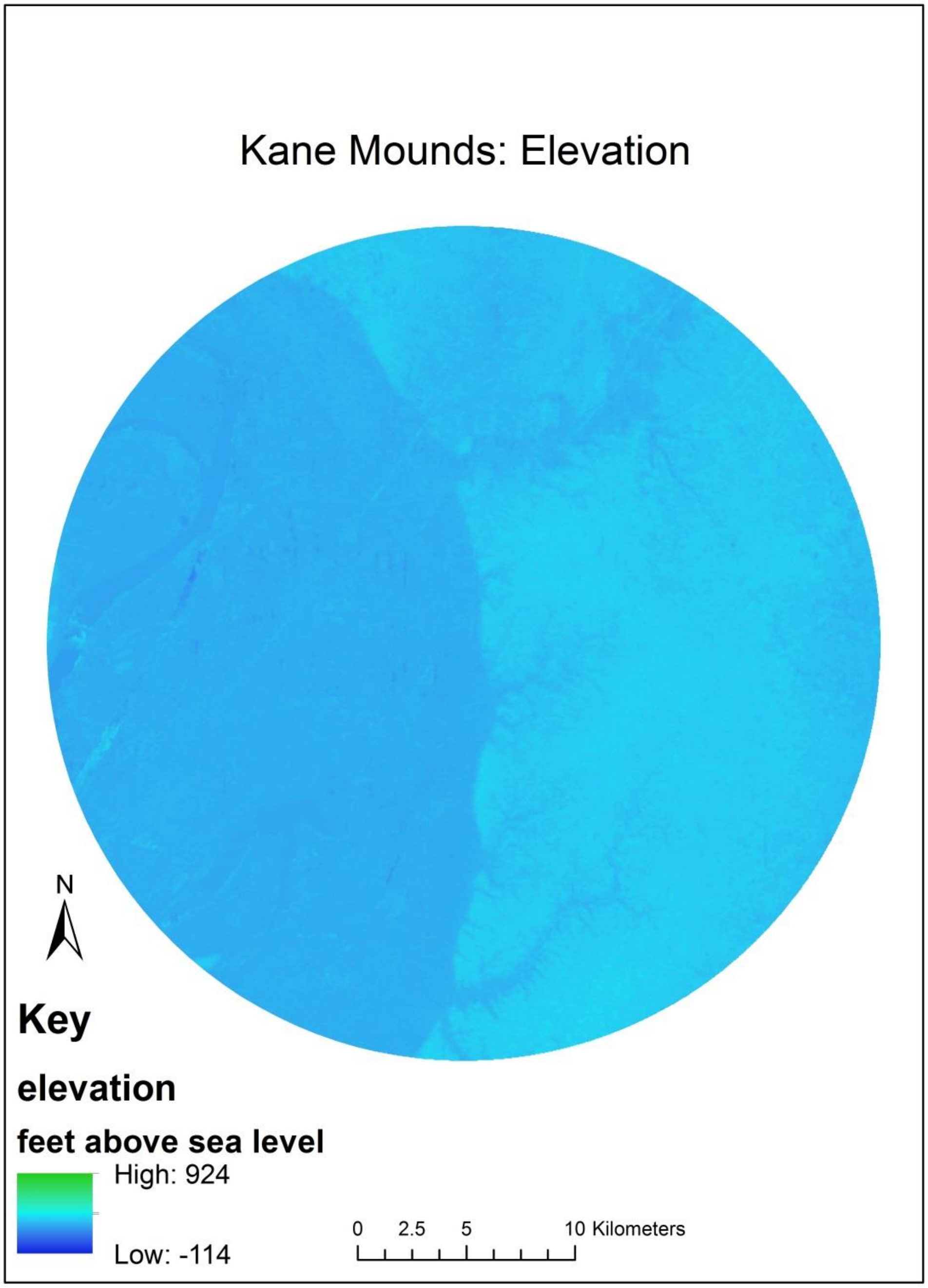

Figure 72. Elevation at Kane Mounds.

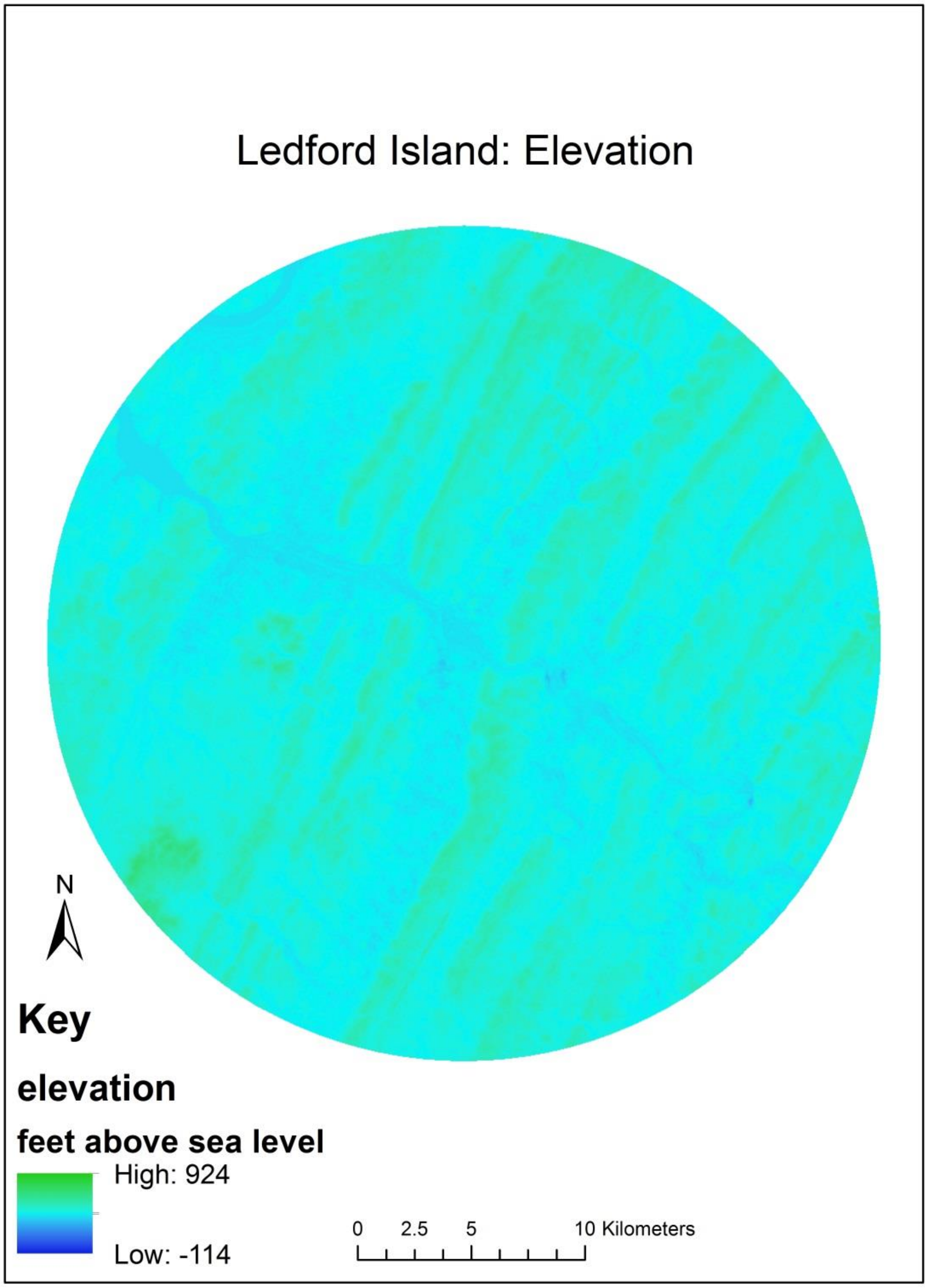

Figure 73. Elevation at Ledford Island.

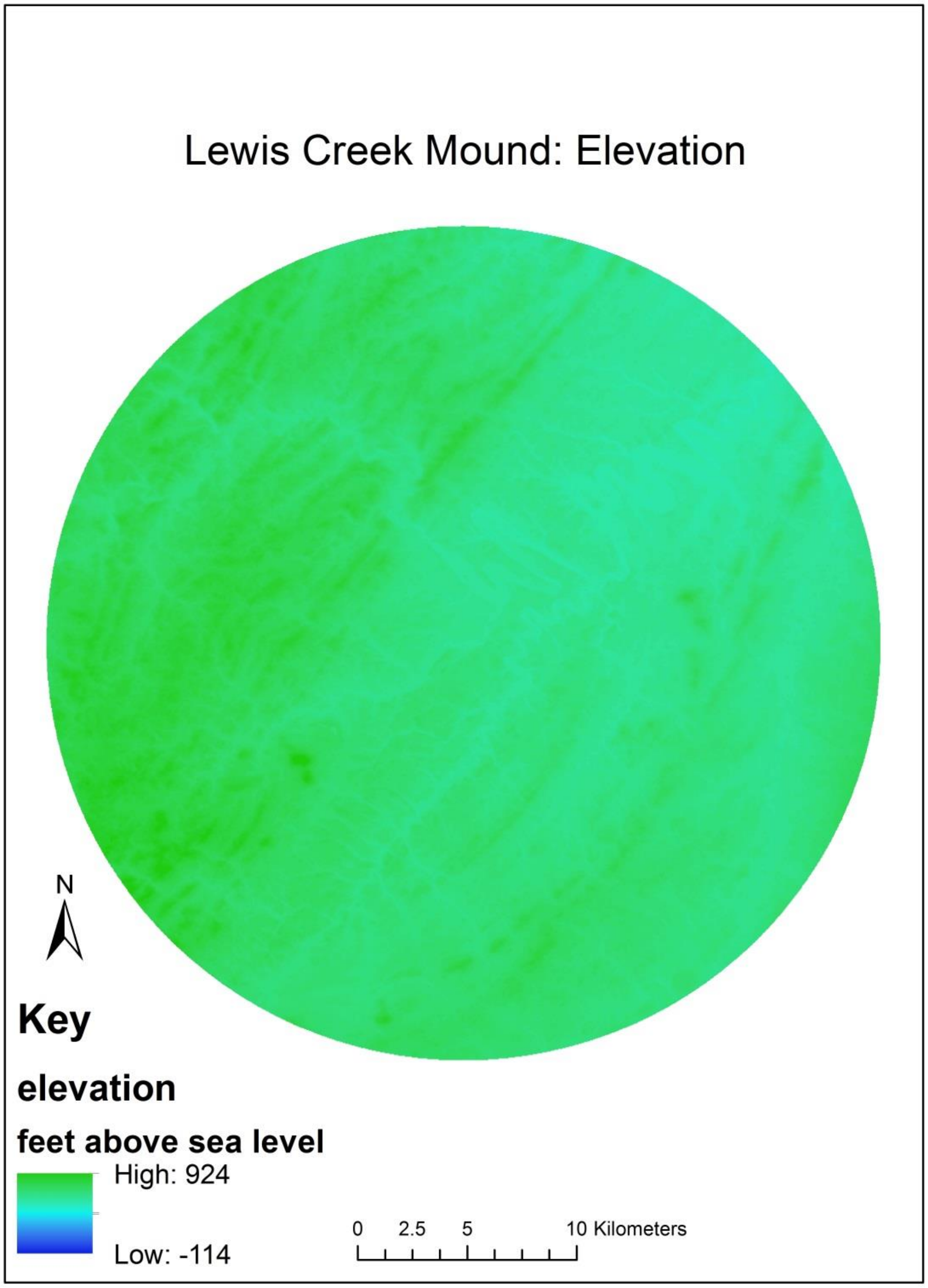

Figure 74. Elevation at Lewis Creek Mound.

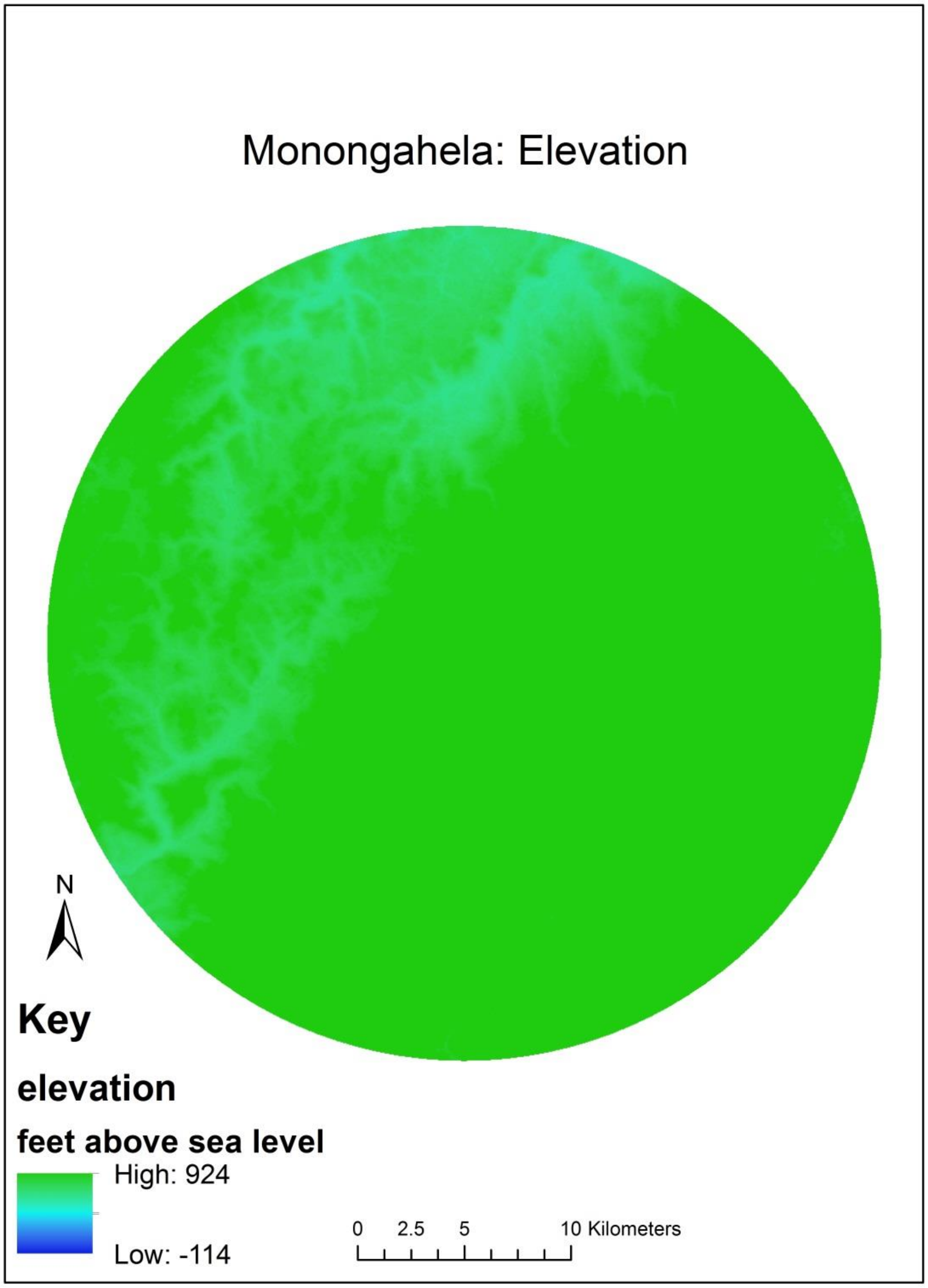

Figure 75. Elevation at Monongahela.

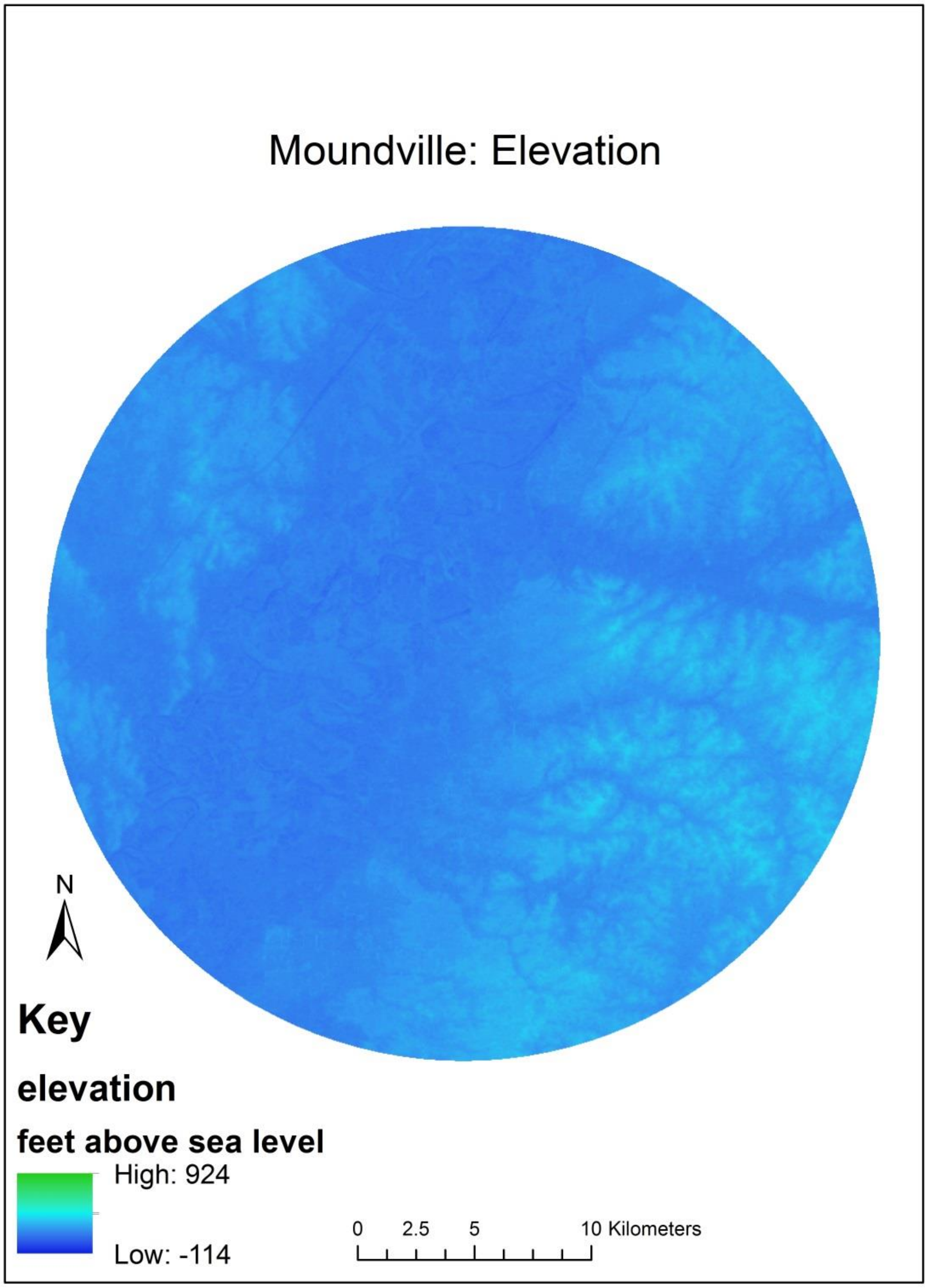

Figure 76. Elevation at Moundville.

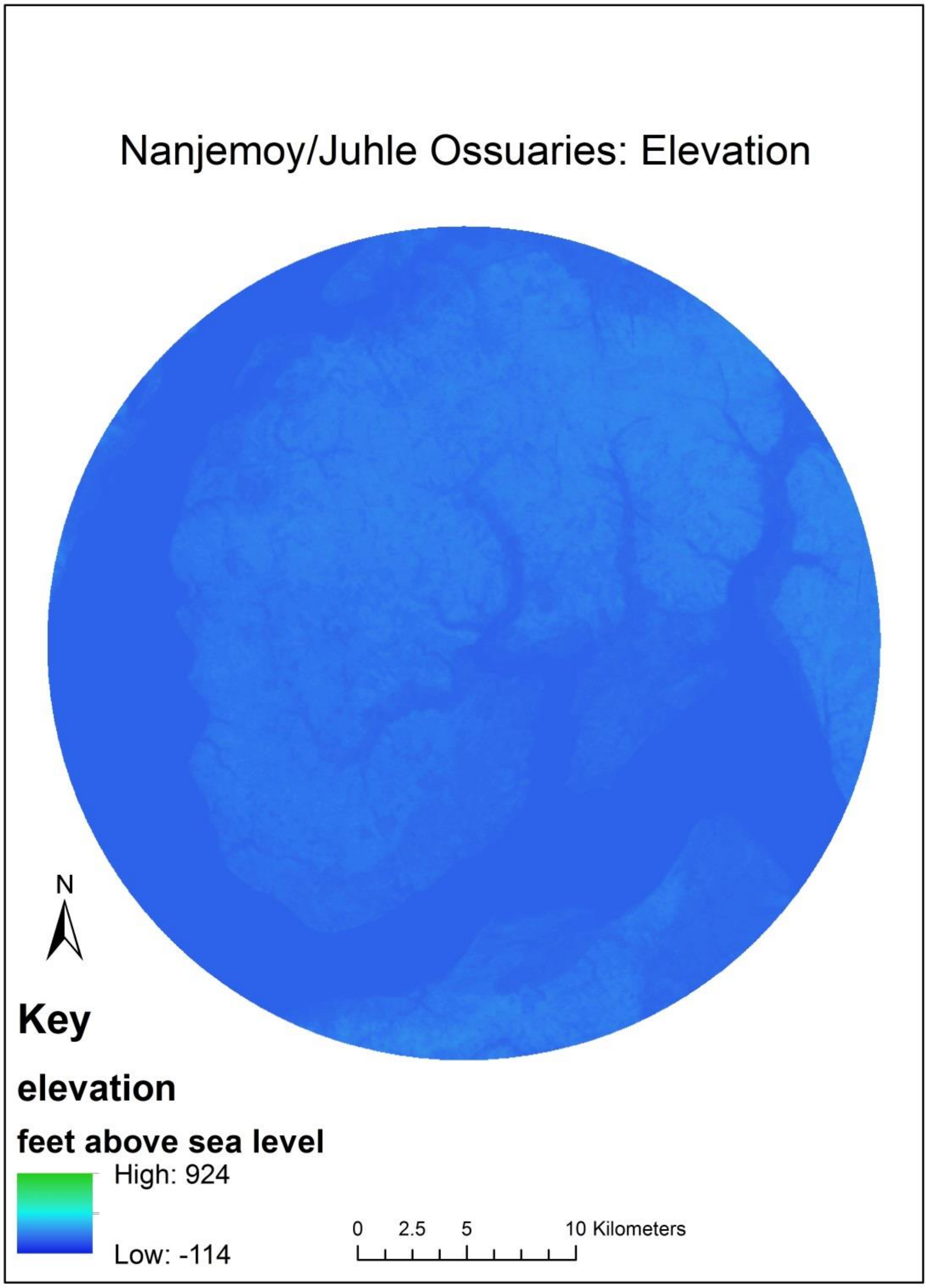

Figure 77. Elevation at Juhle Ossuary.

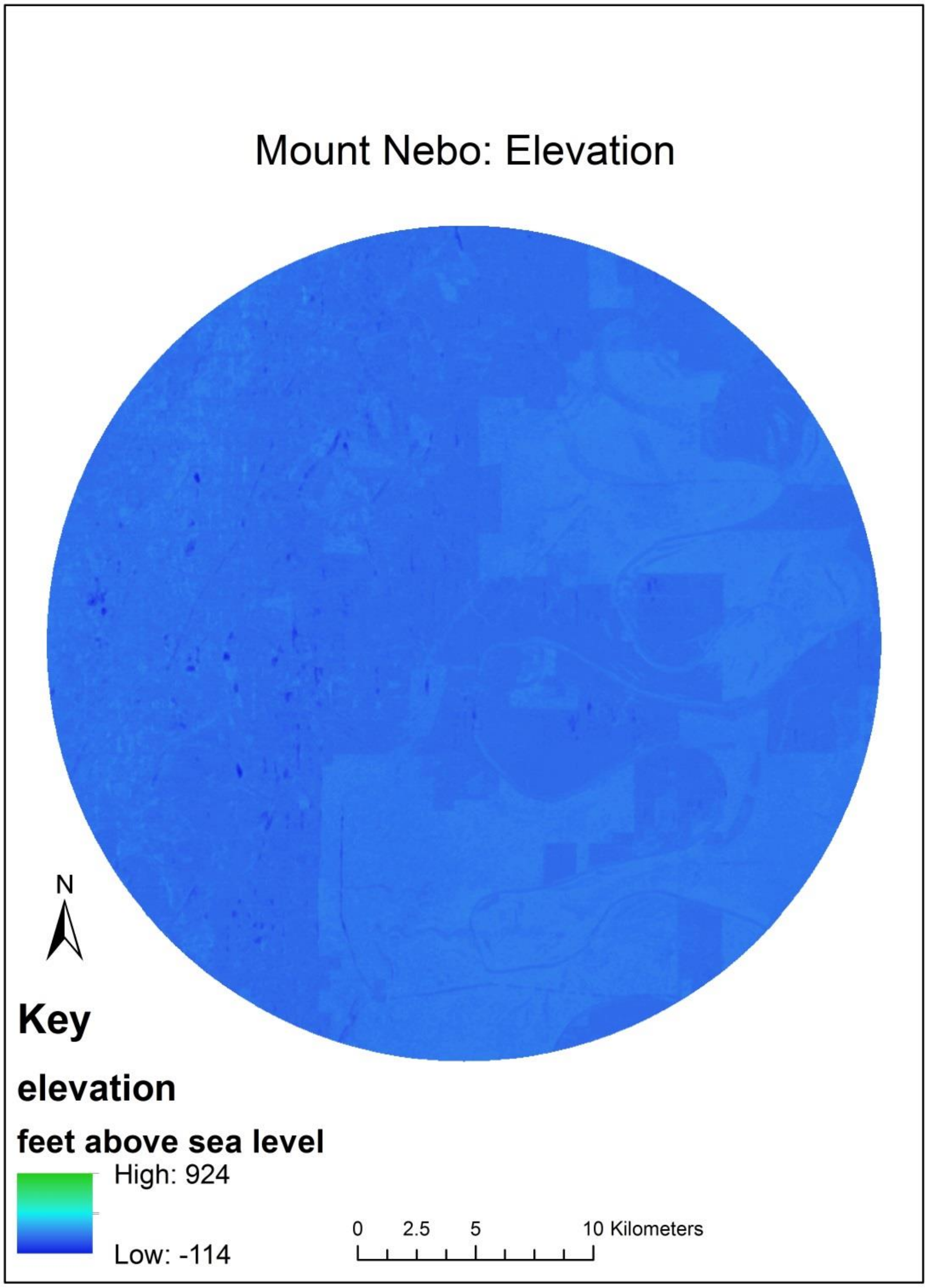

Figure 78. Elevation at Mount Nebo.

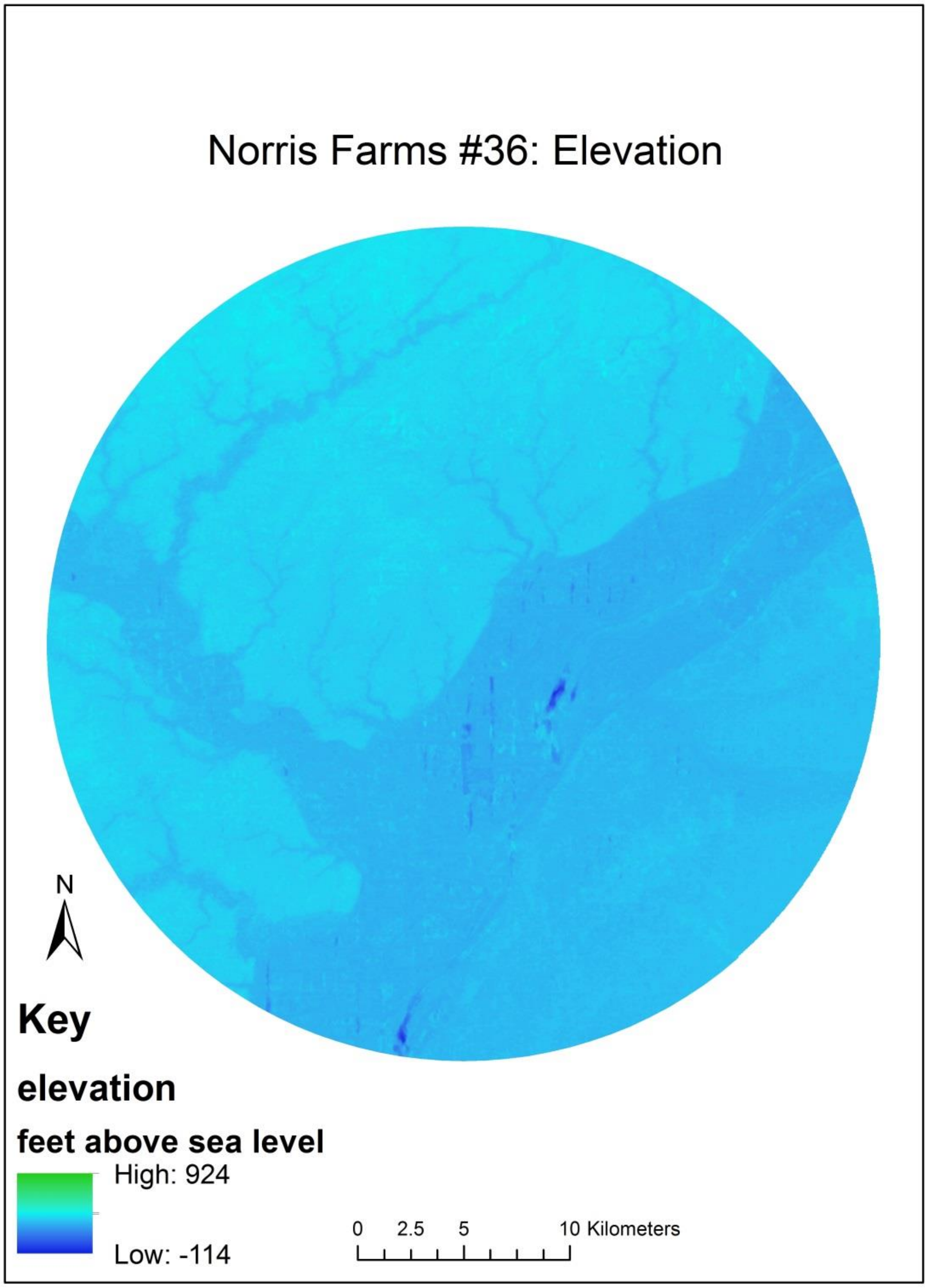

Figure 79. Elevation at Norris Farms #36.

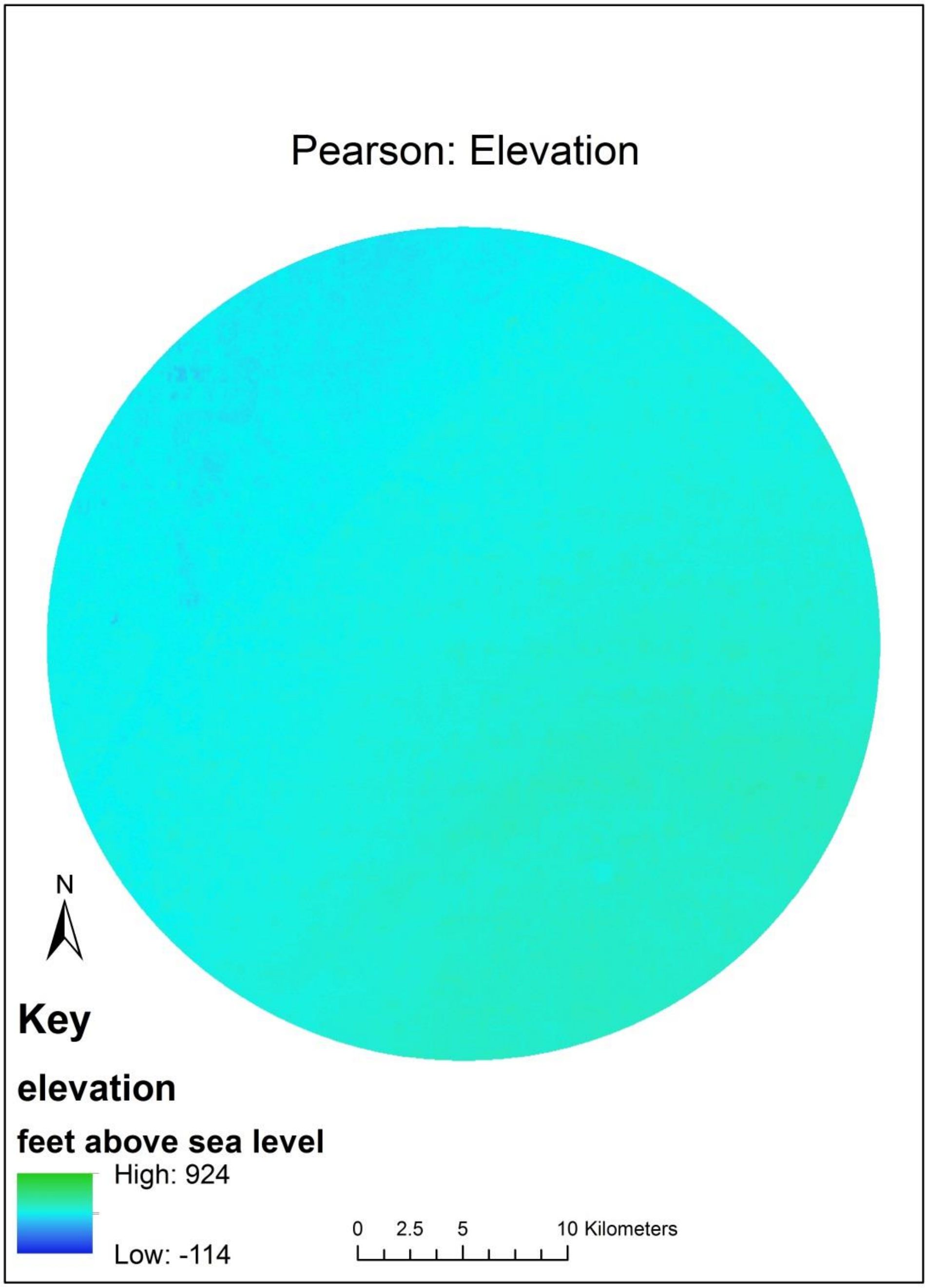

Figure 80. Elevation at Pearson.

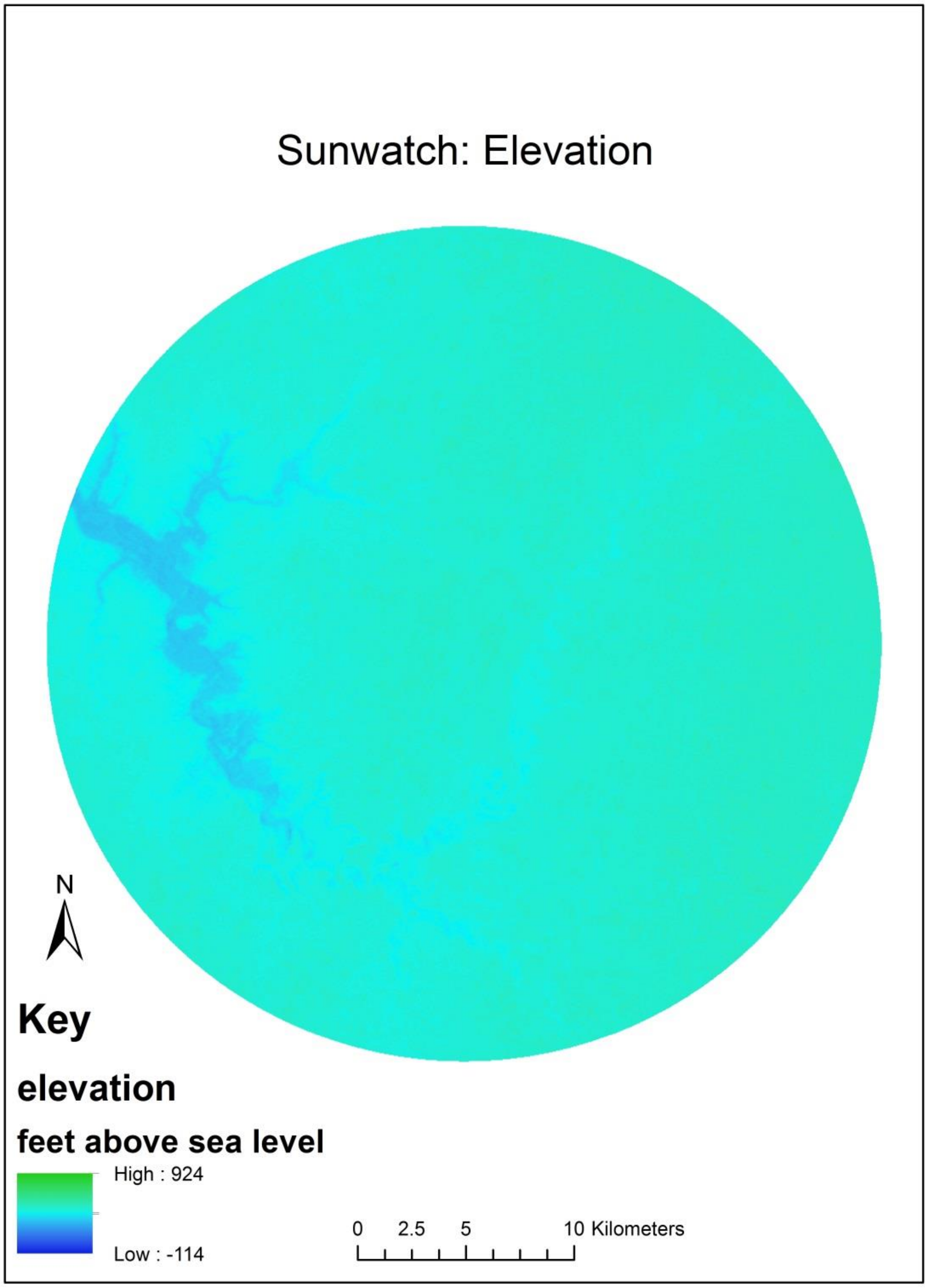

Figure 81. Elevation at Sunwatch.

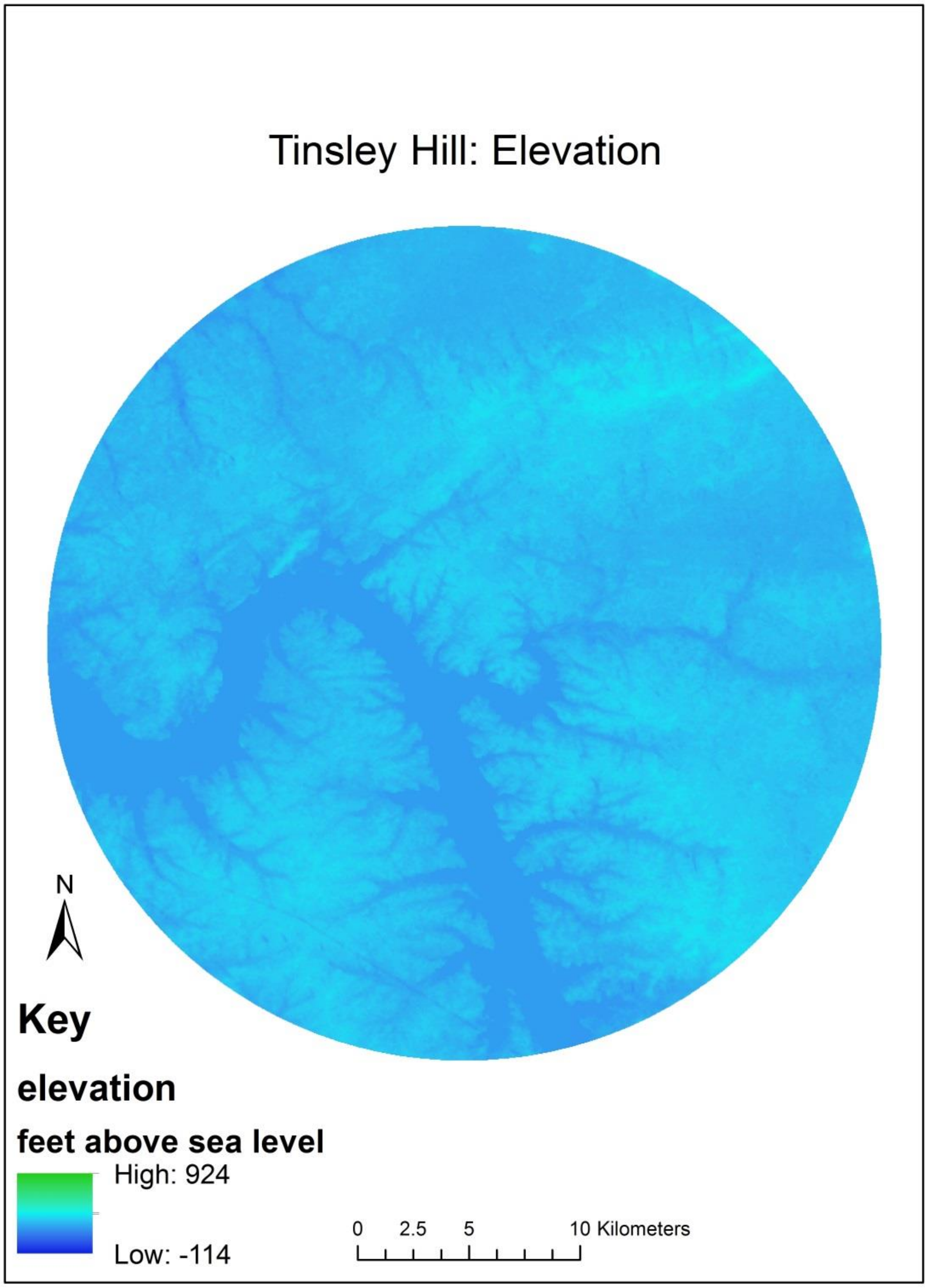

Figure 82. Elevation at Tinsley Hill.

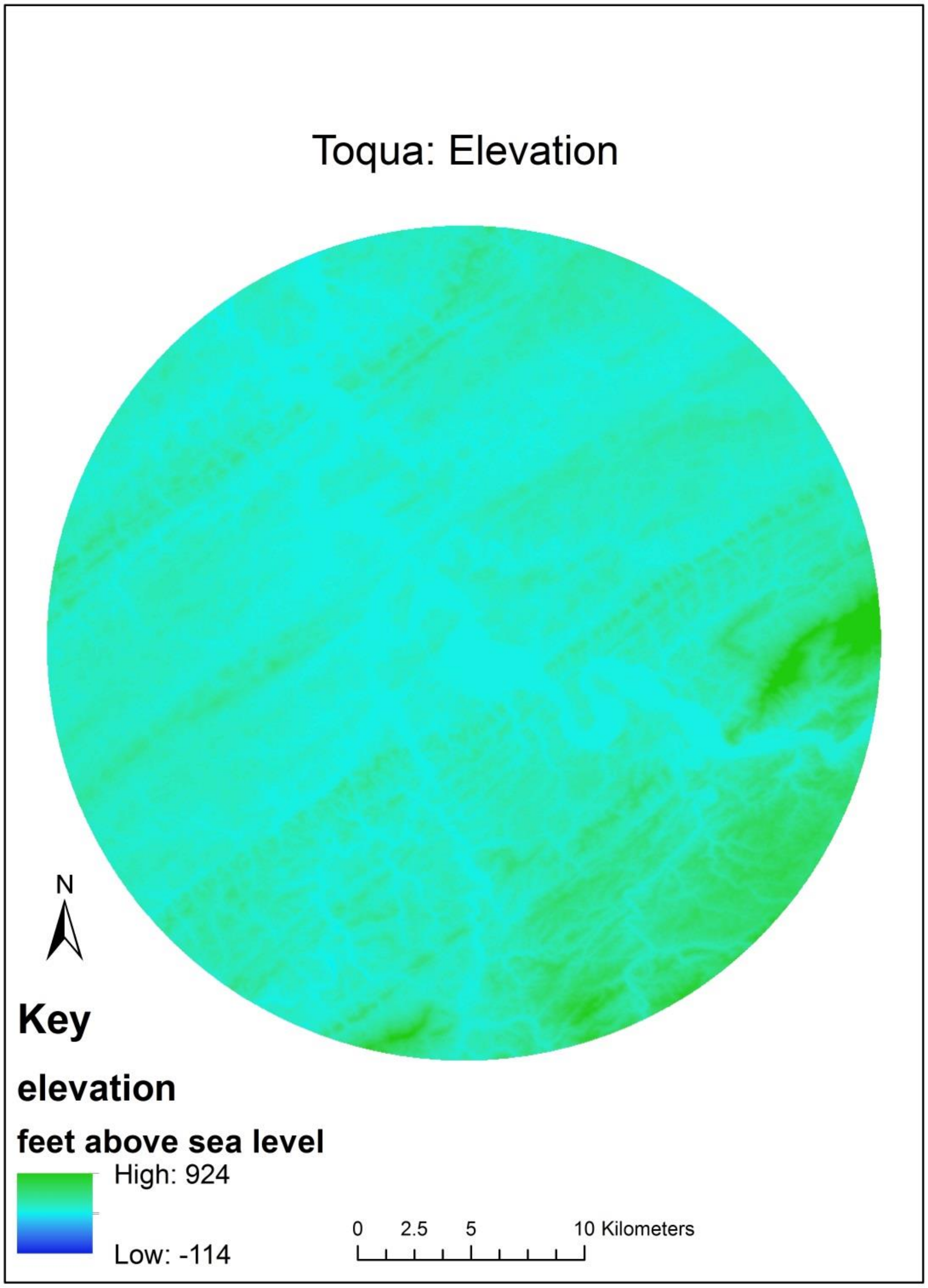

Figure 83. Elevation at Toqua.

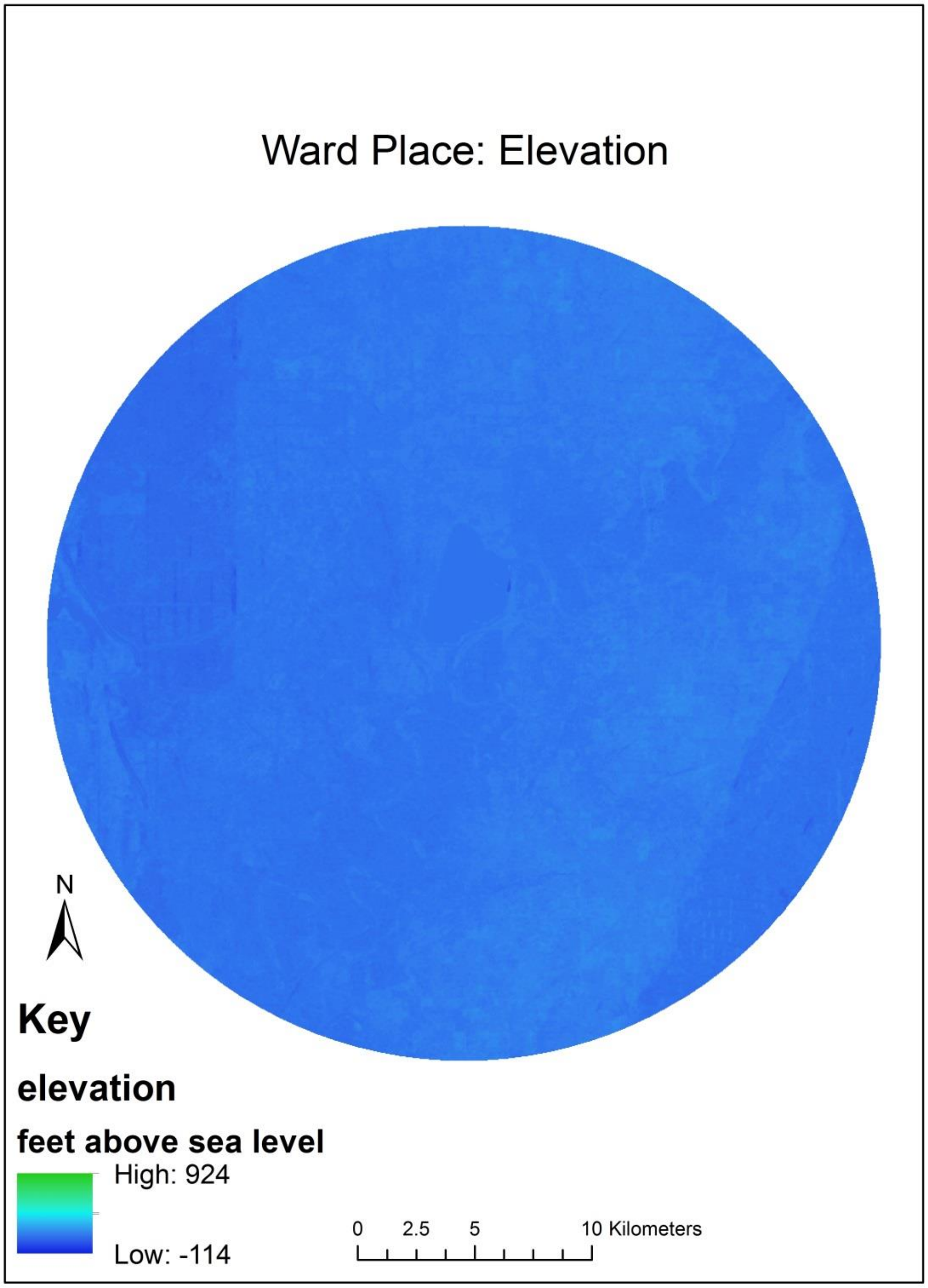

Figure 84. Elevation at Ward Place.

A4. Precipitation

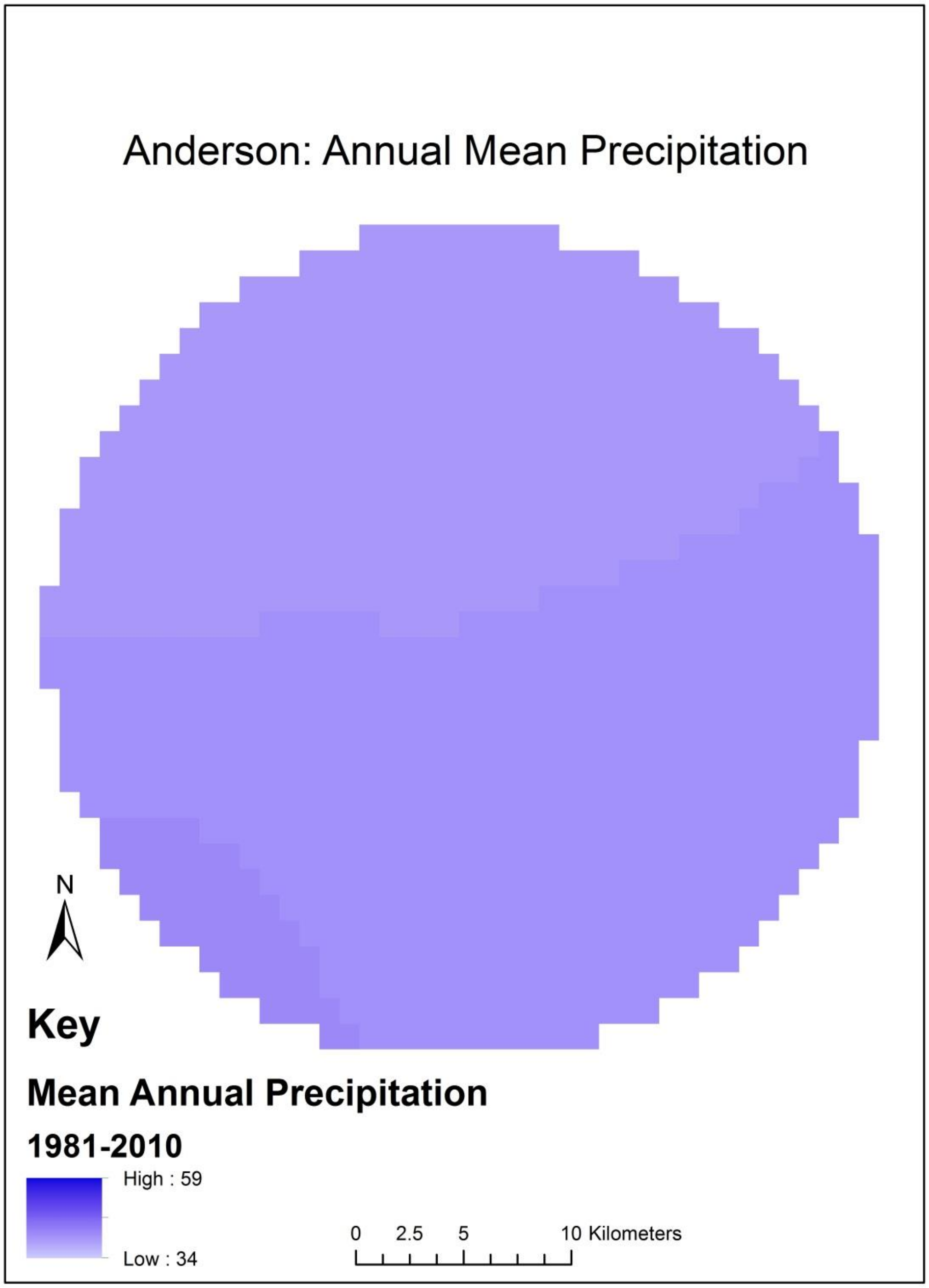

Figure 85. Precipitation at Anderson.

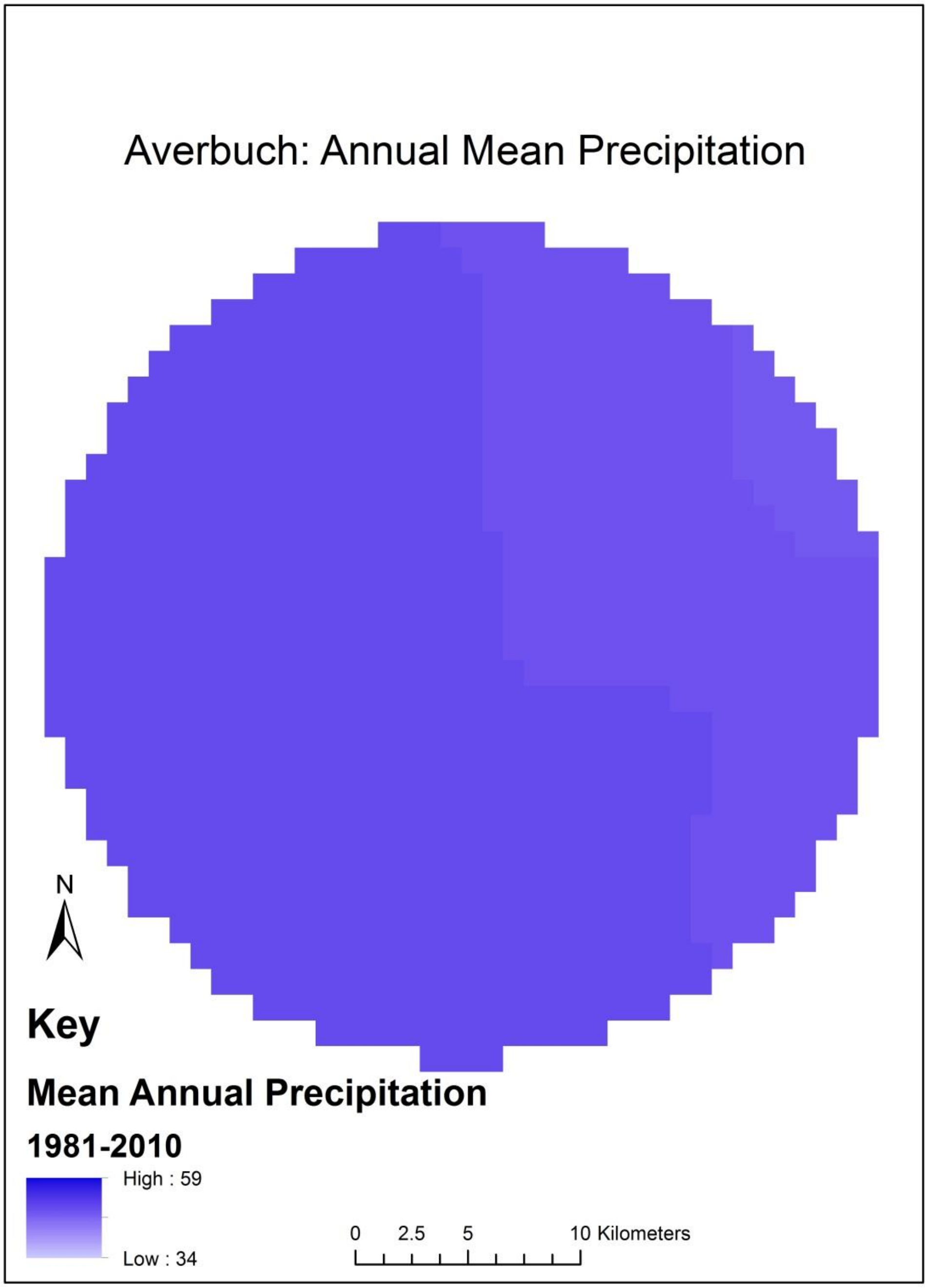

Figure 86. Precipitation at Averbuch.

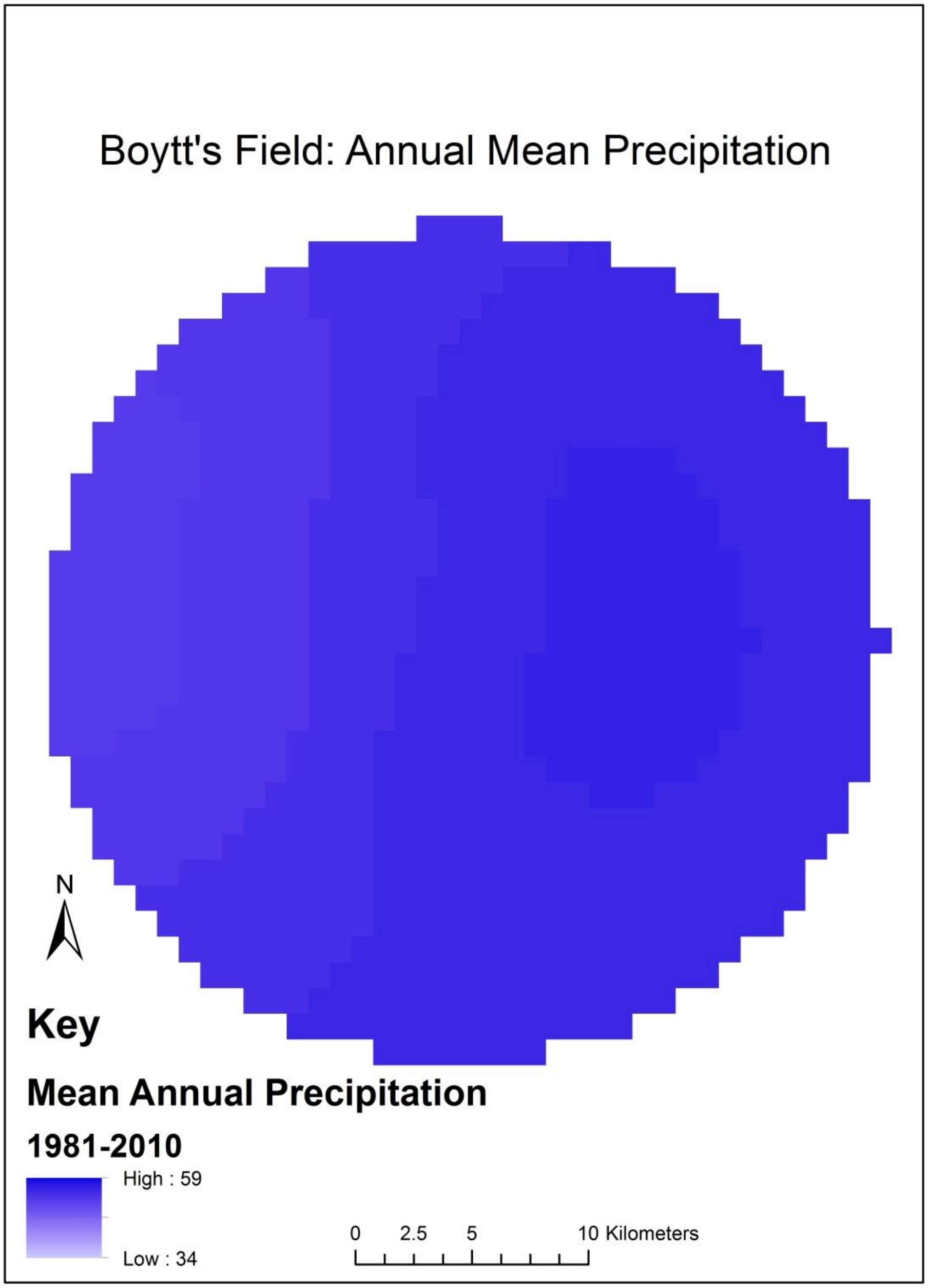

Figure 87. Precipitation at Boytt's Field.

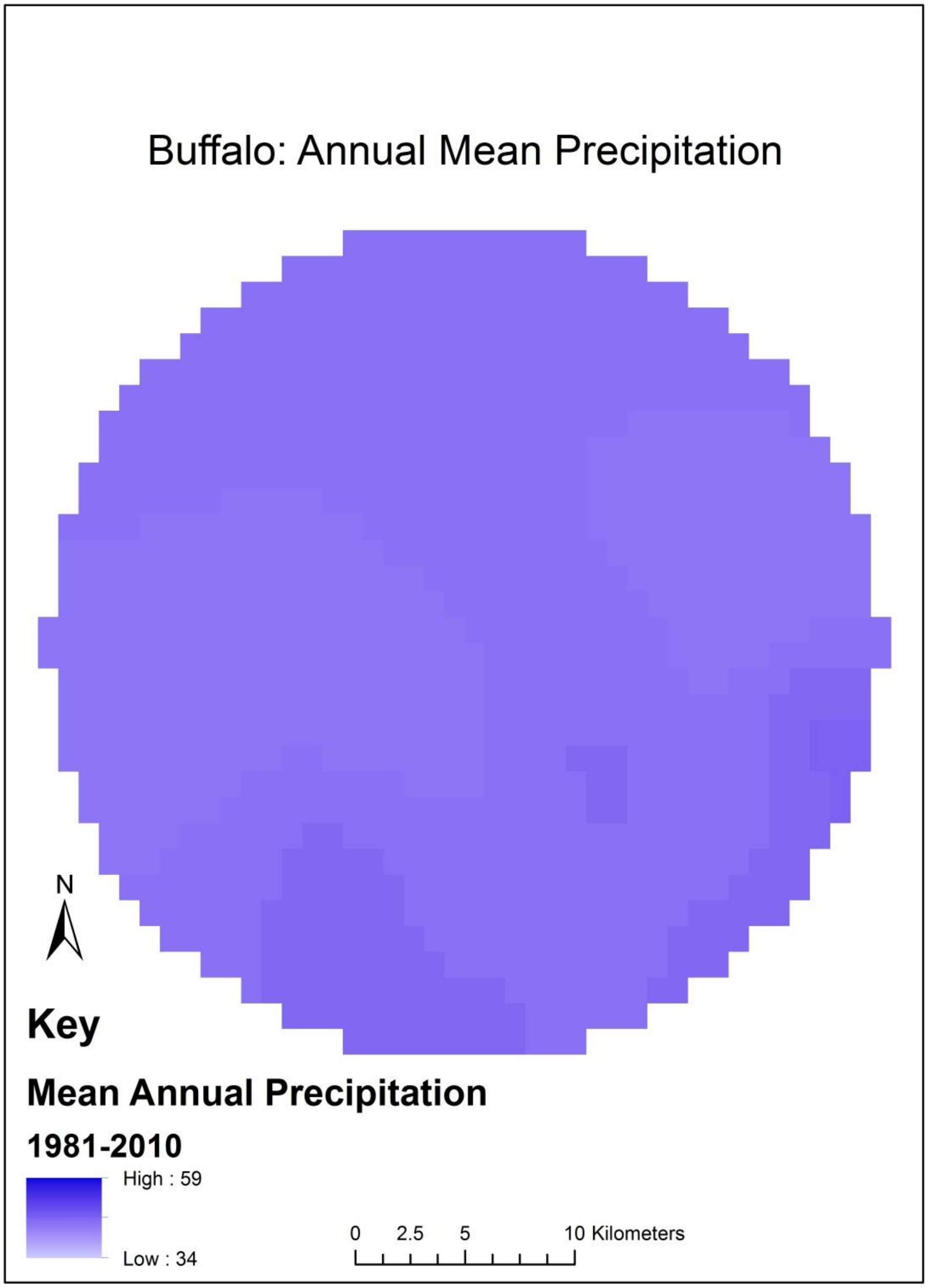

Figure 88. Precipitation at Buffalo.

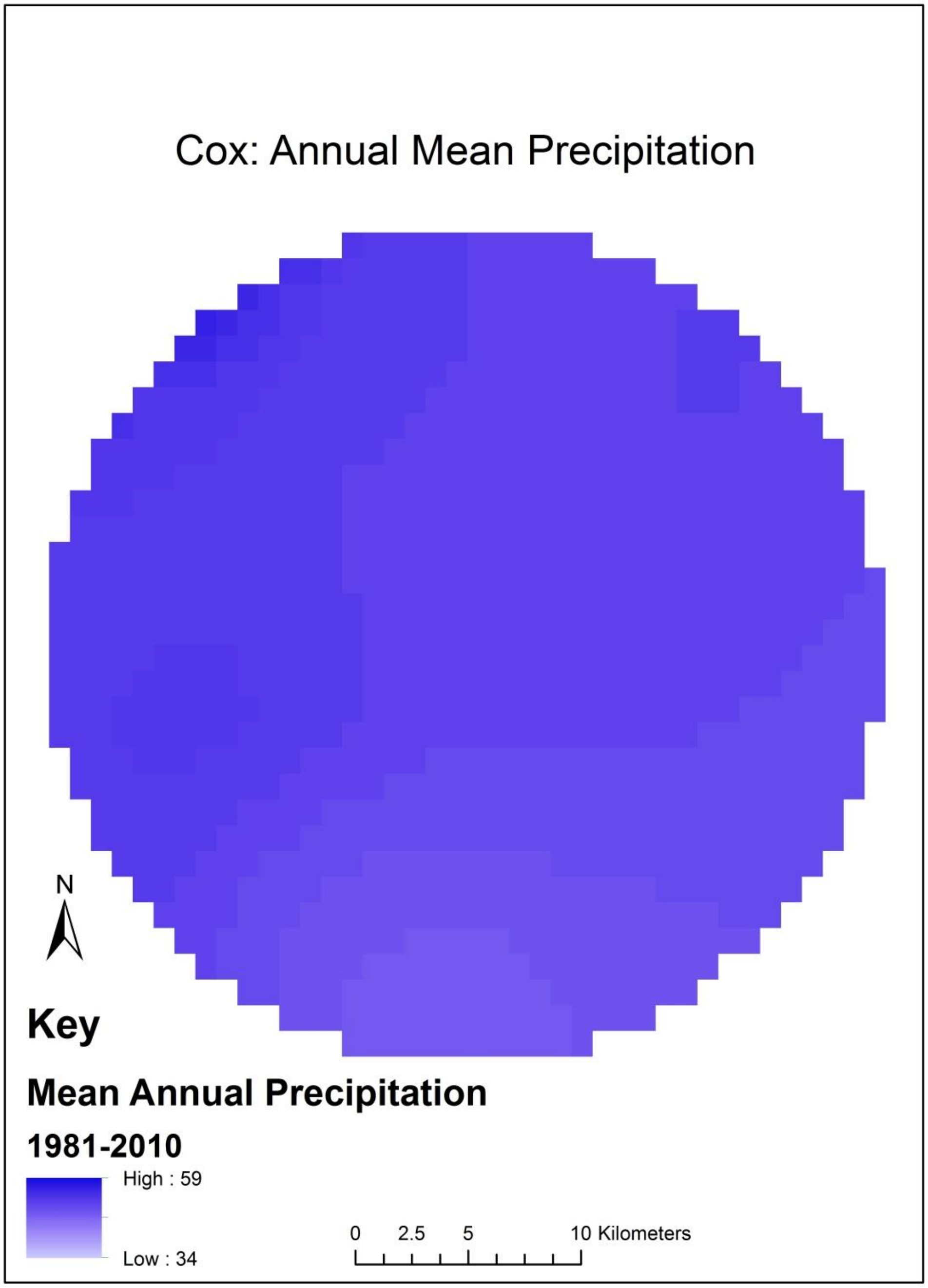

Figure 89. Precipitation at Cox.

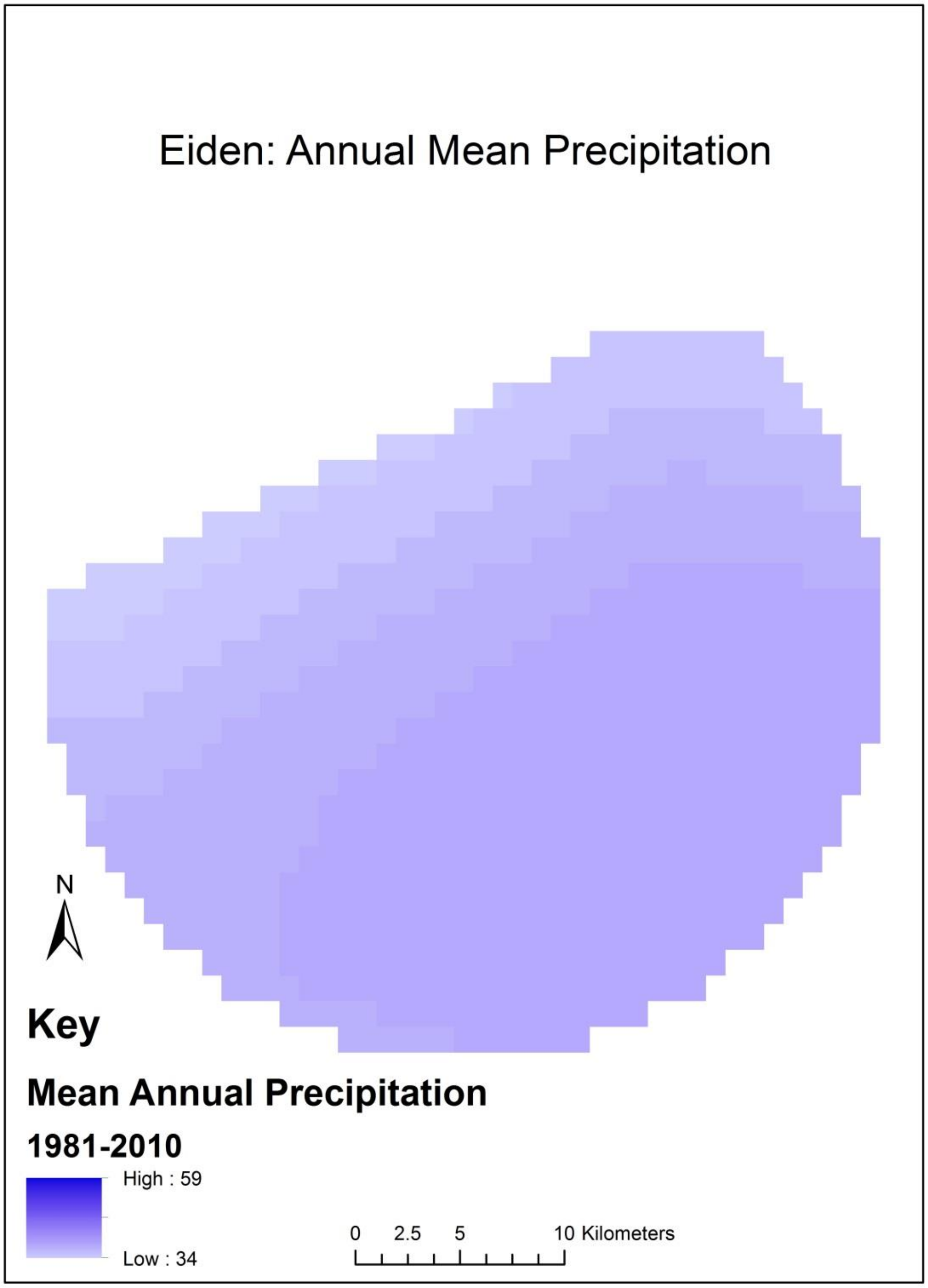

Figure 90. Precipitation at Eiden.

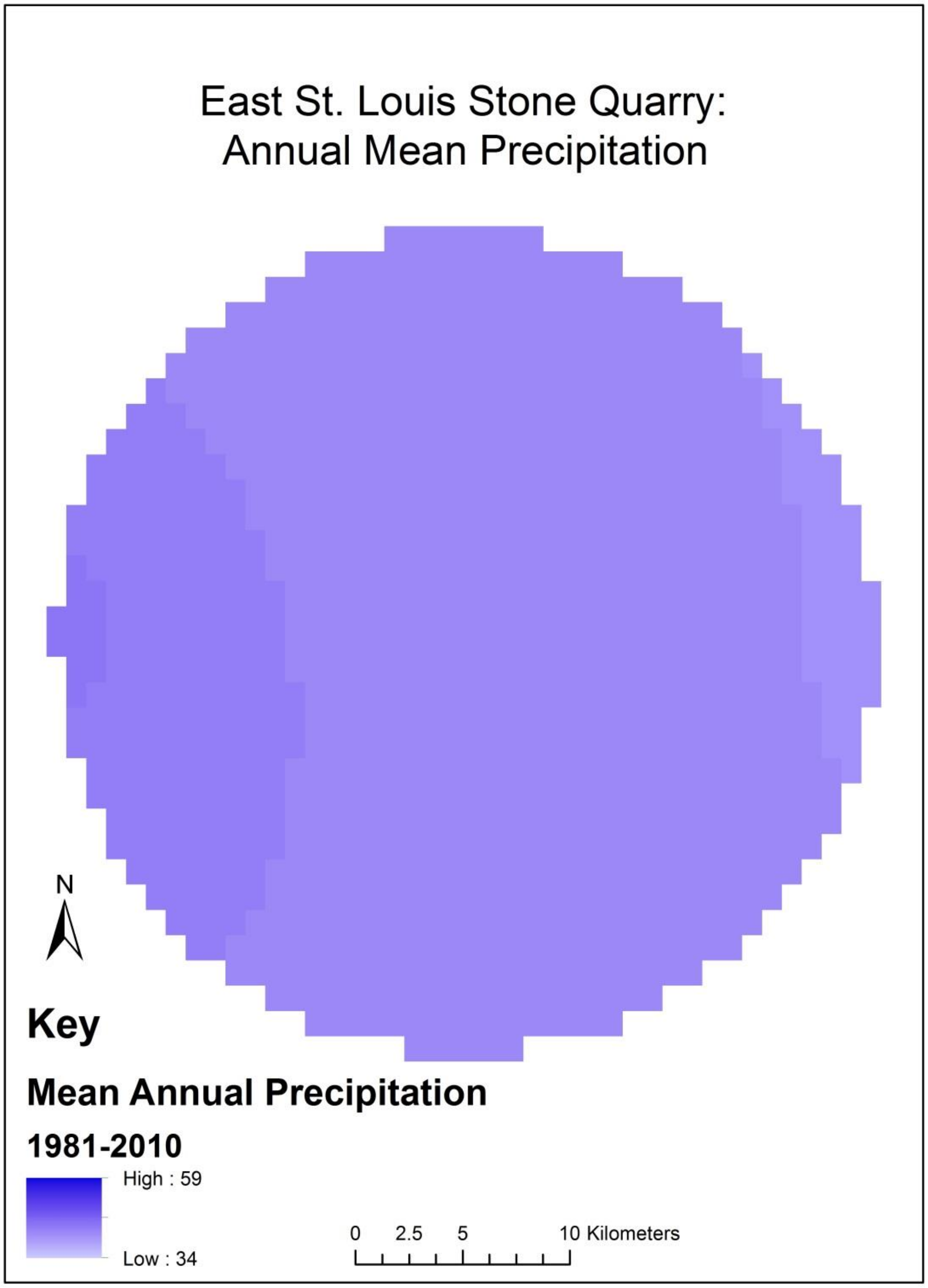

Figure 91. Precipitation at East St. Louis Stone Quarry.

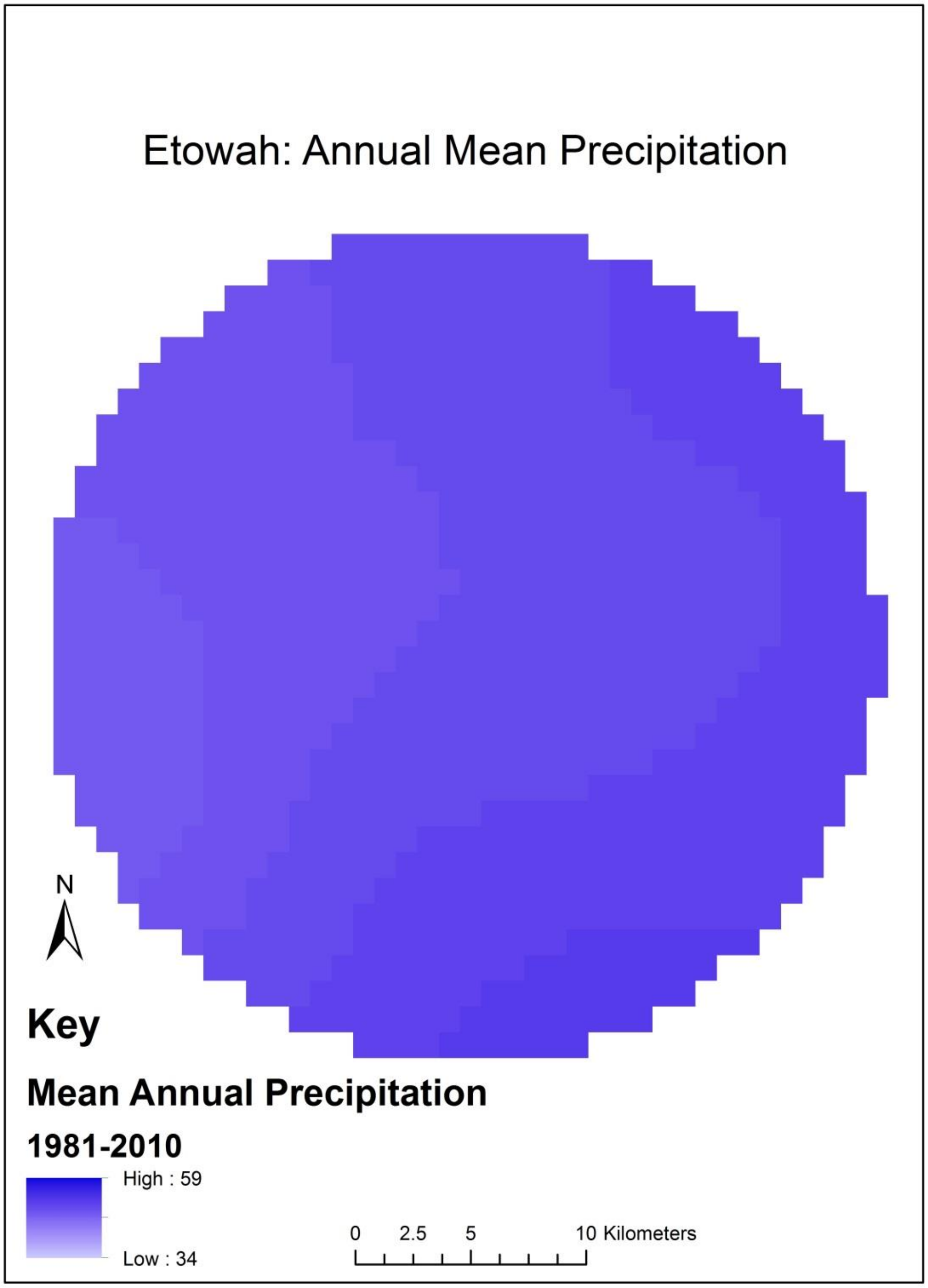

Figure 92. Precipitation at Etowah.

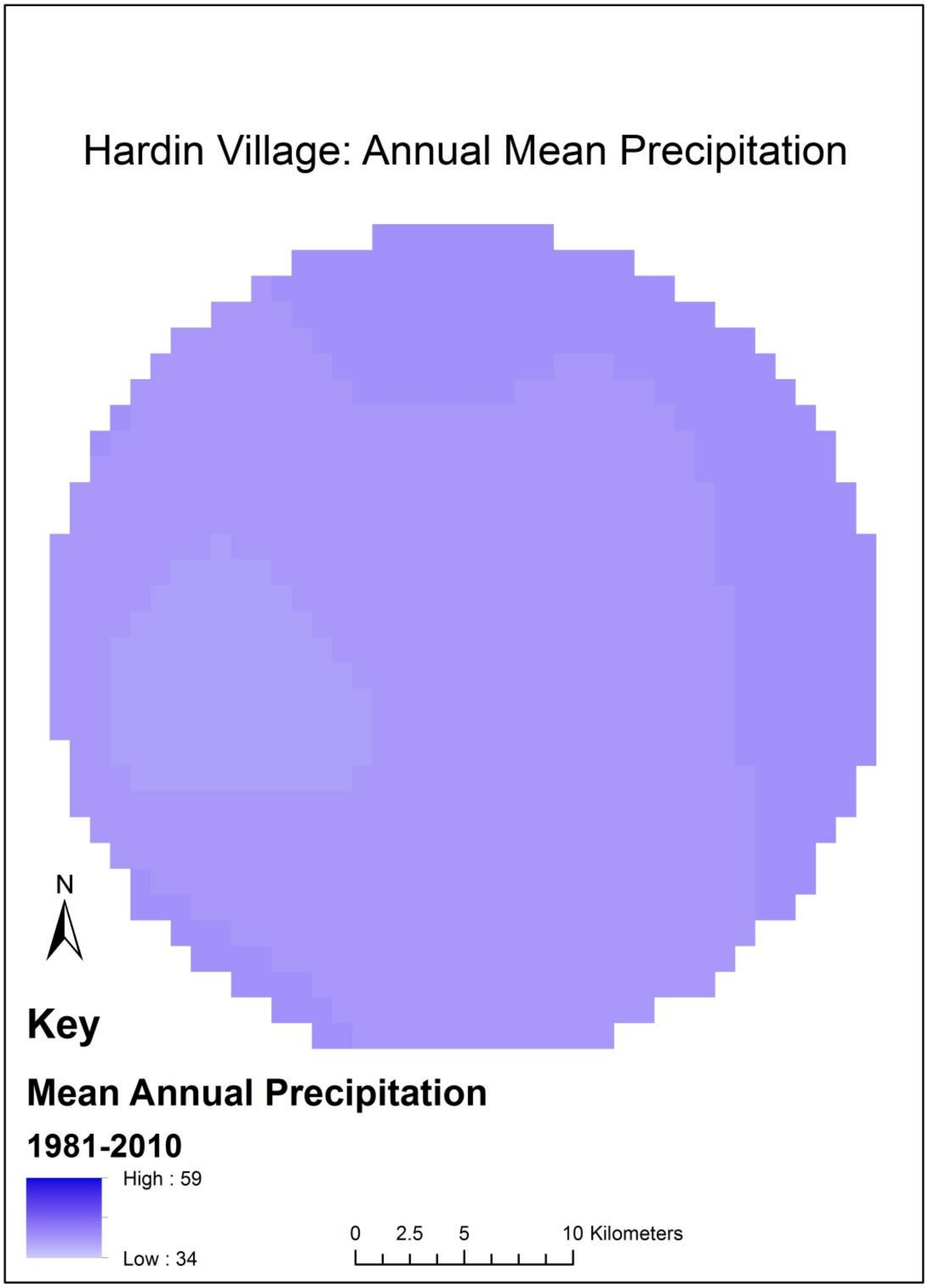

Figure 93. Precipitation at Hardin Village.

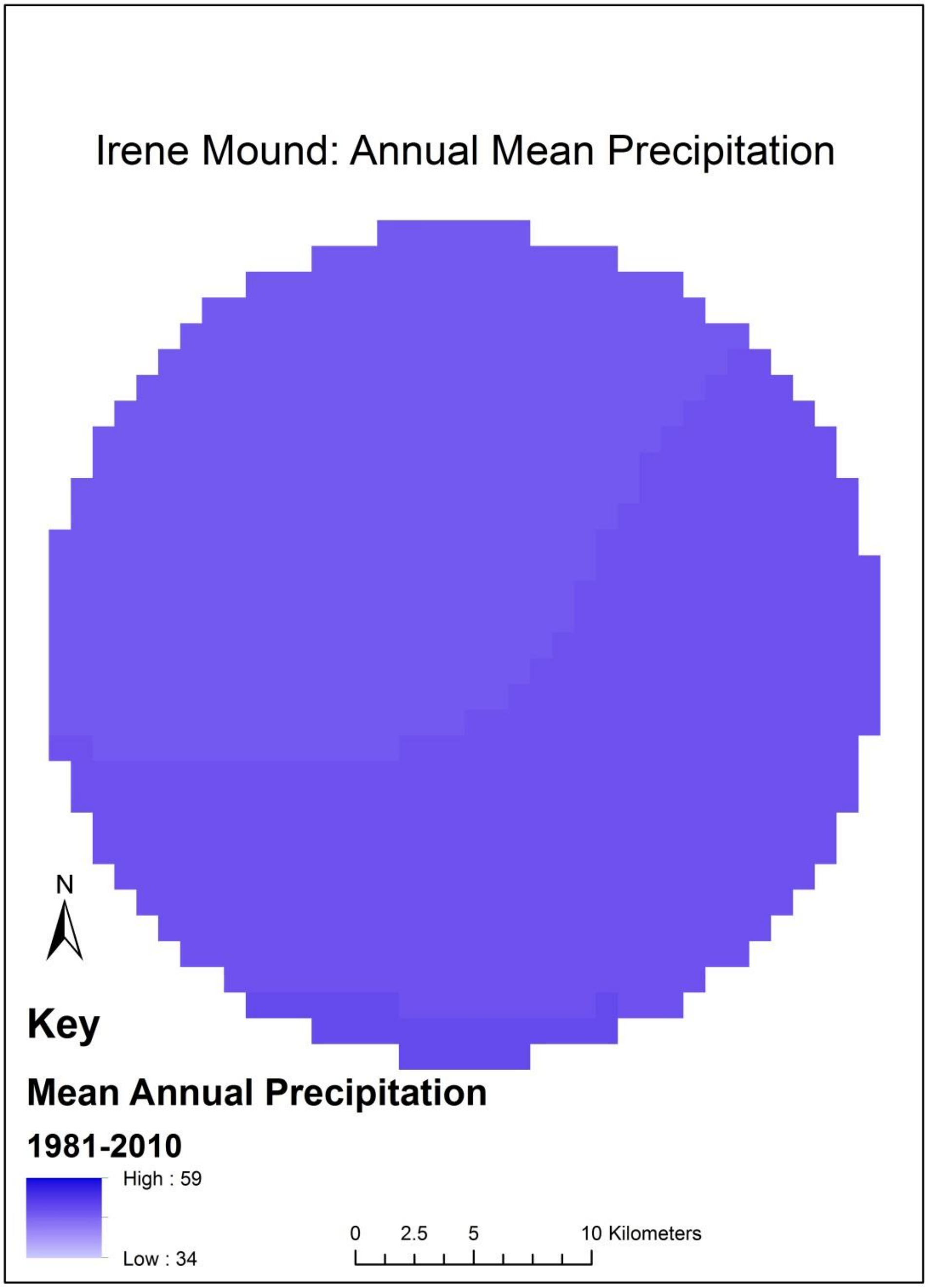

Figure 94. Precipitation at Irene Mound.

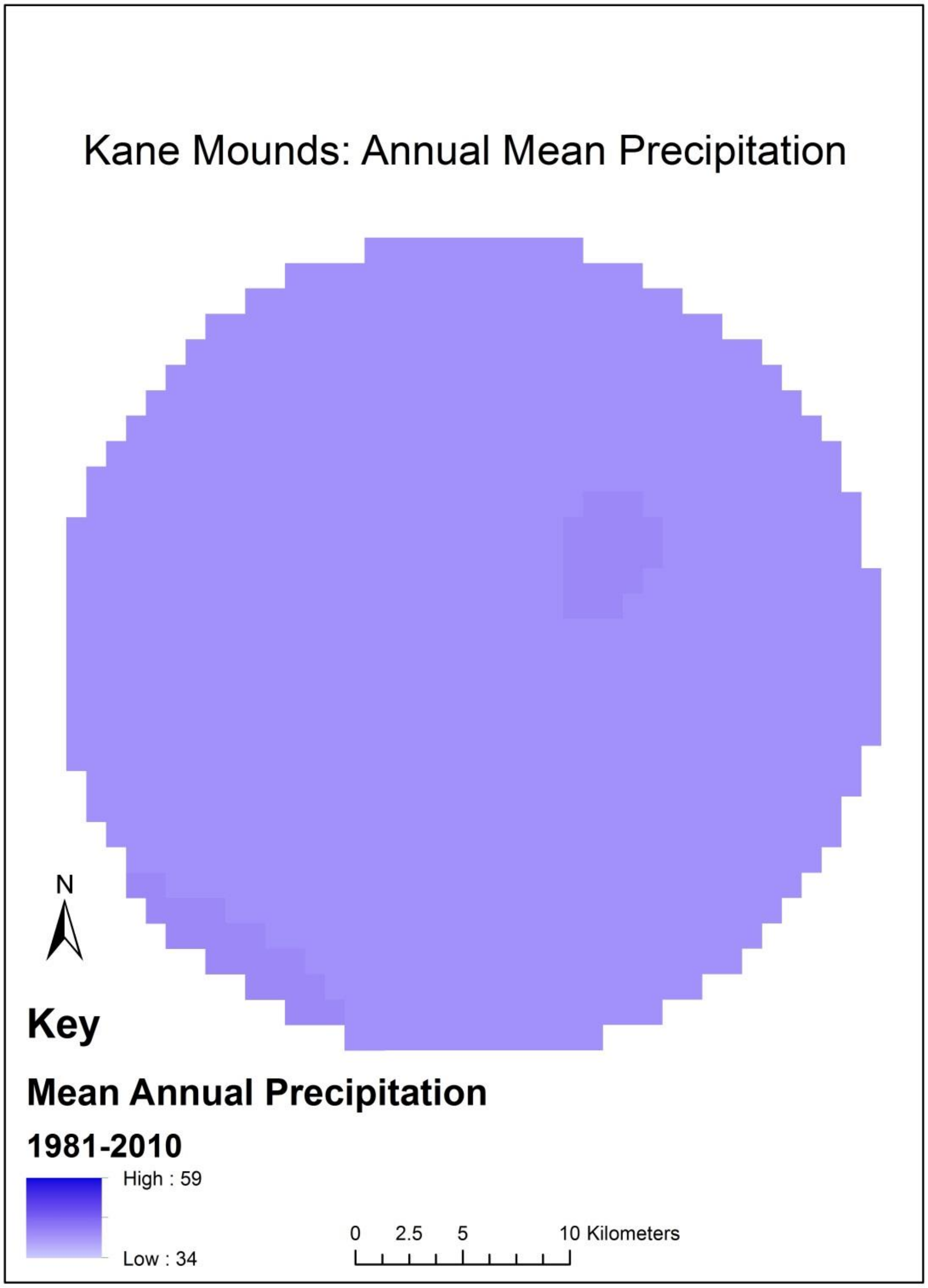

Figure 95. Precipitation at Kane Mounds.

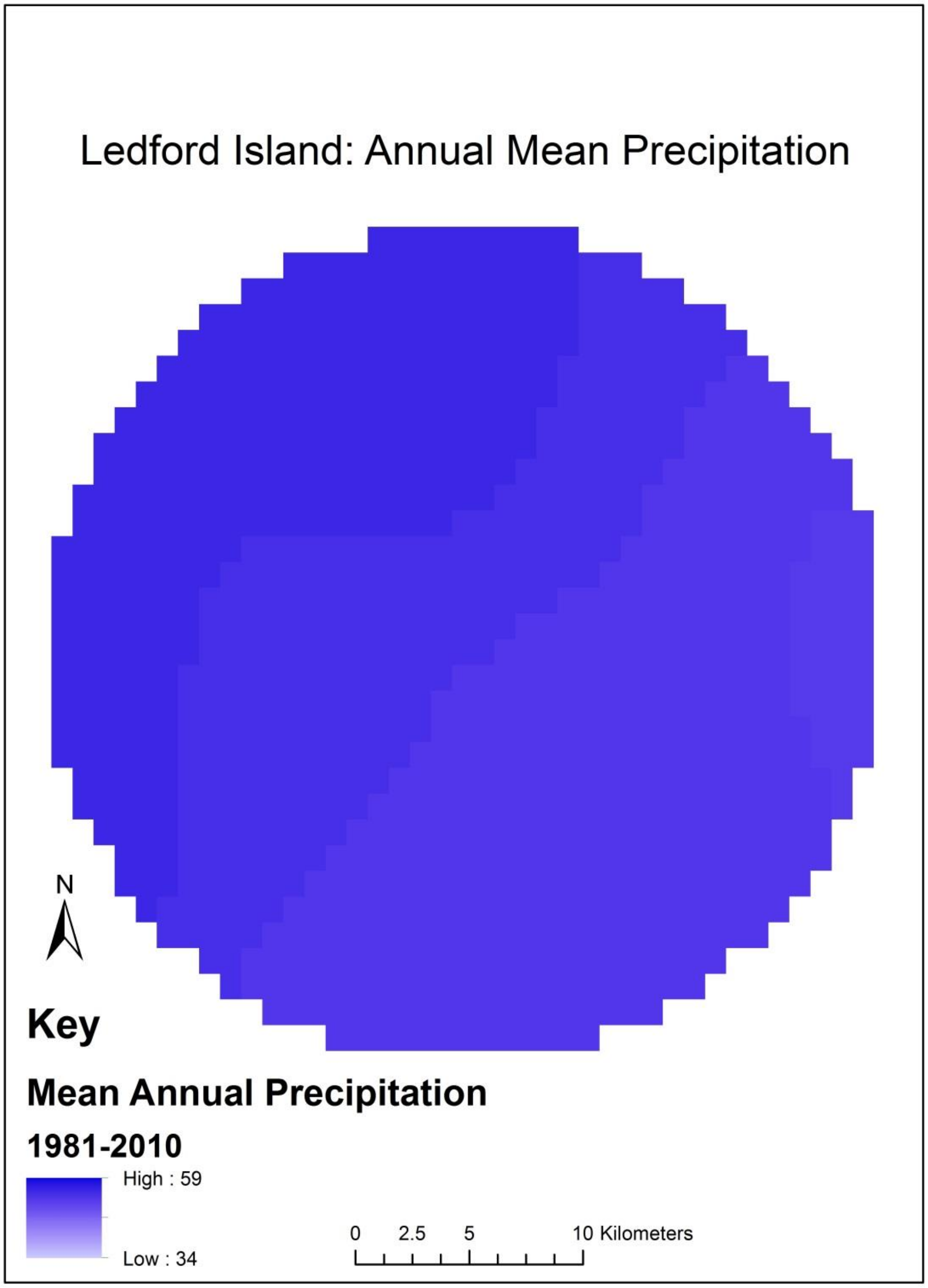

Figure 96. Precipitation at Ledford Island.

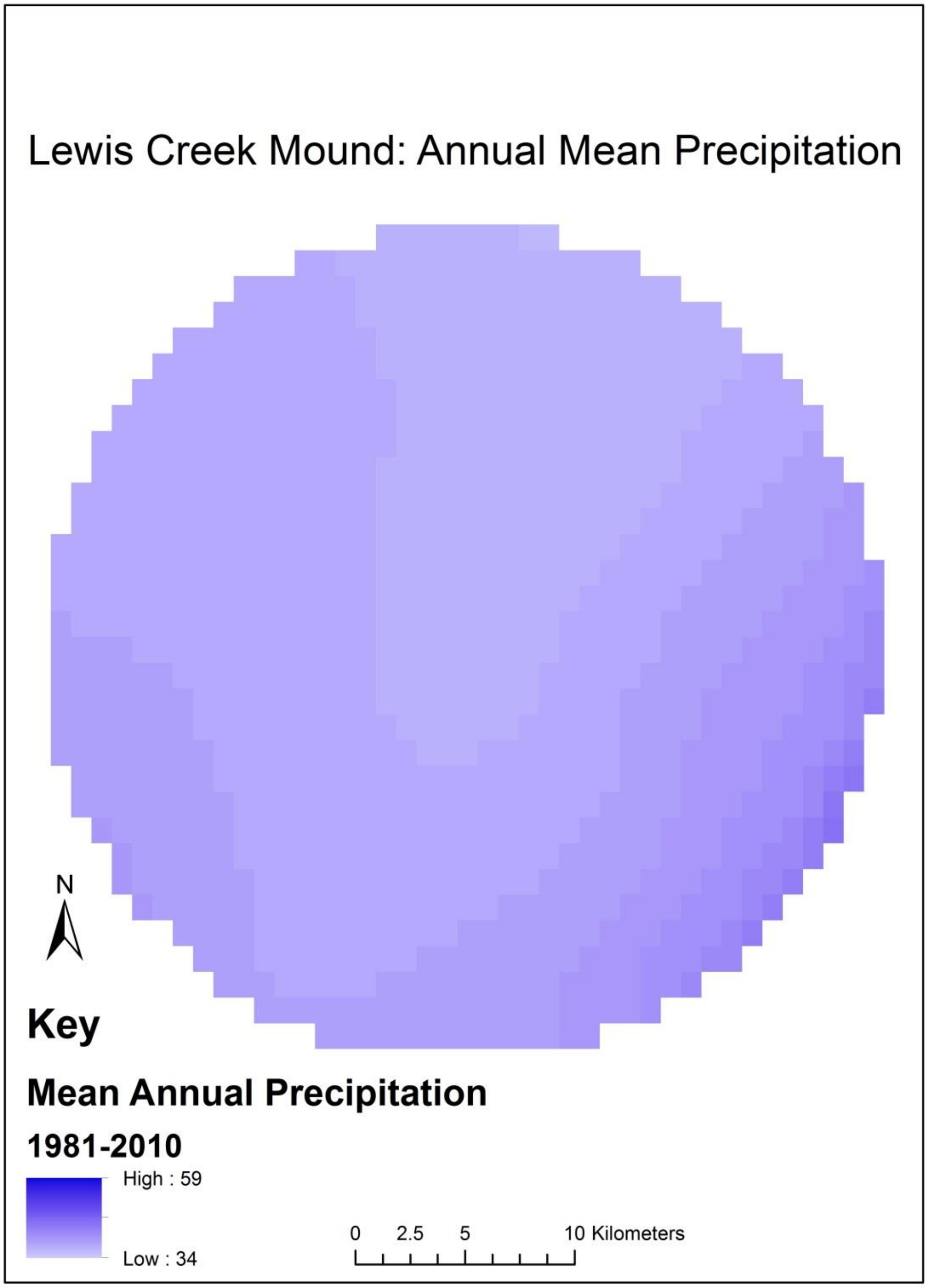

Figure 97. Precipitation at Lewis Creek Mound.

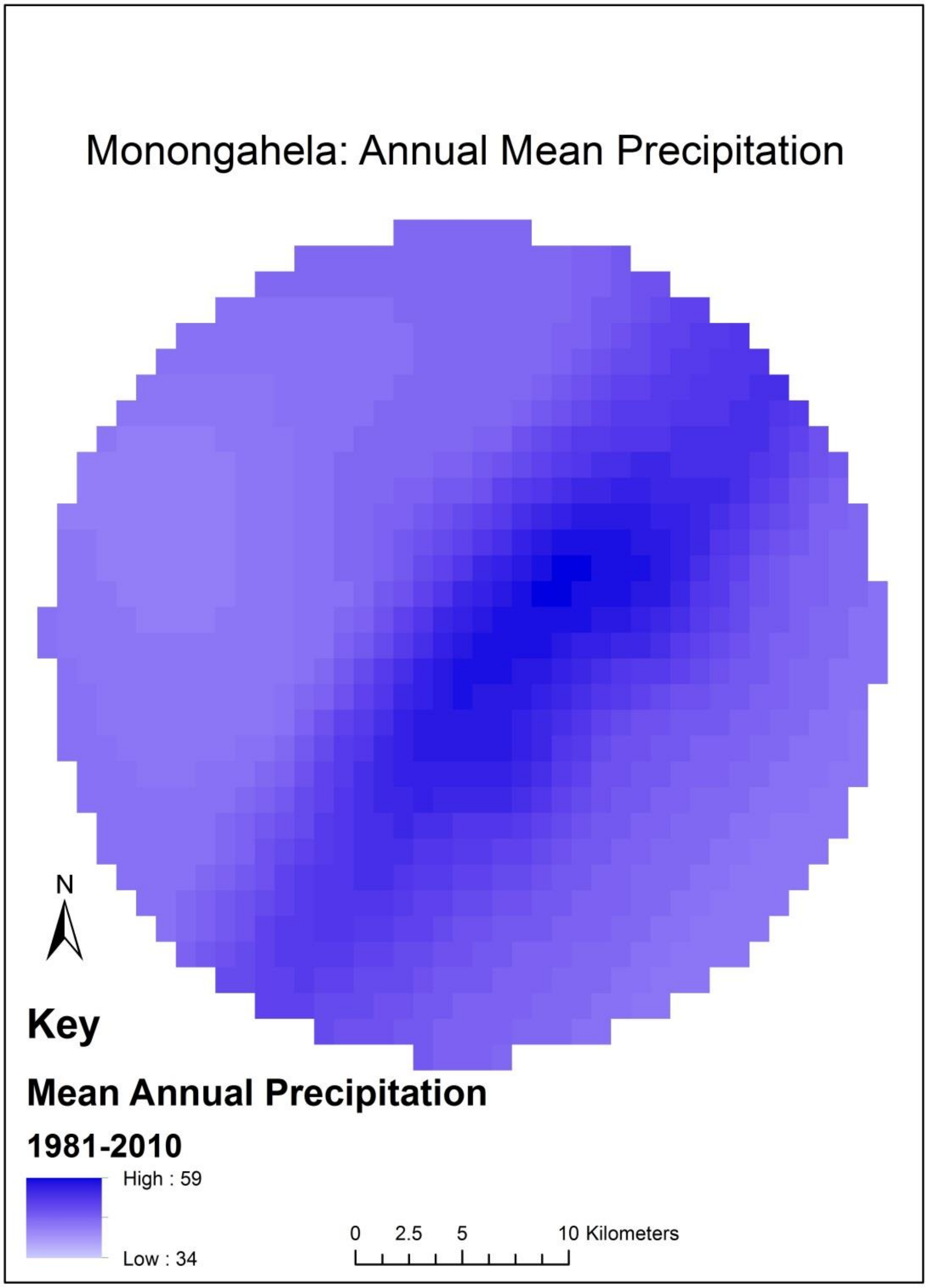

Figure 98. Precipitation at Monongahela.

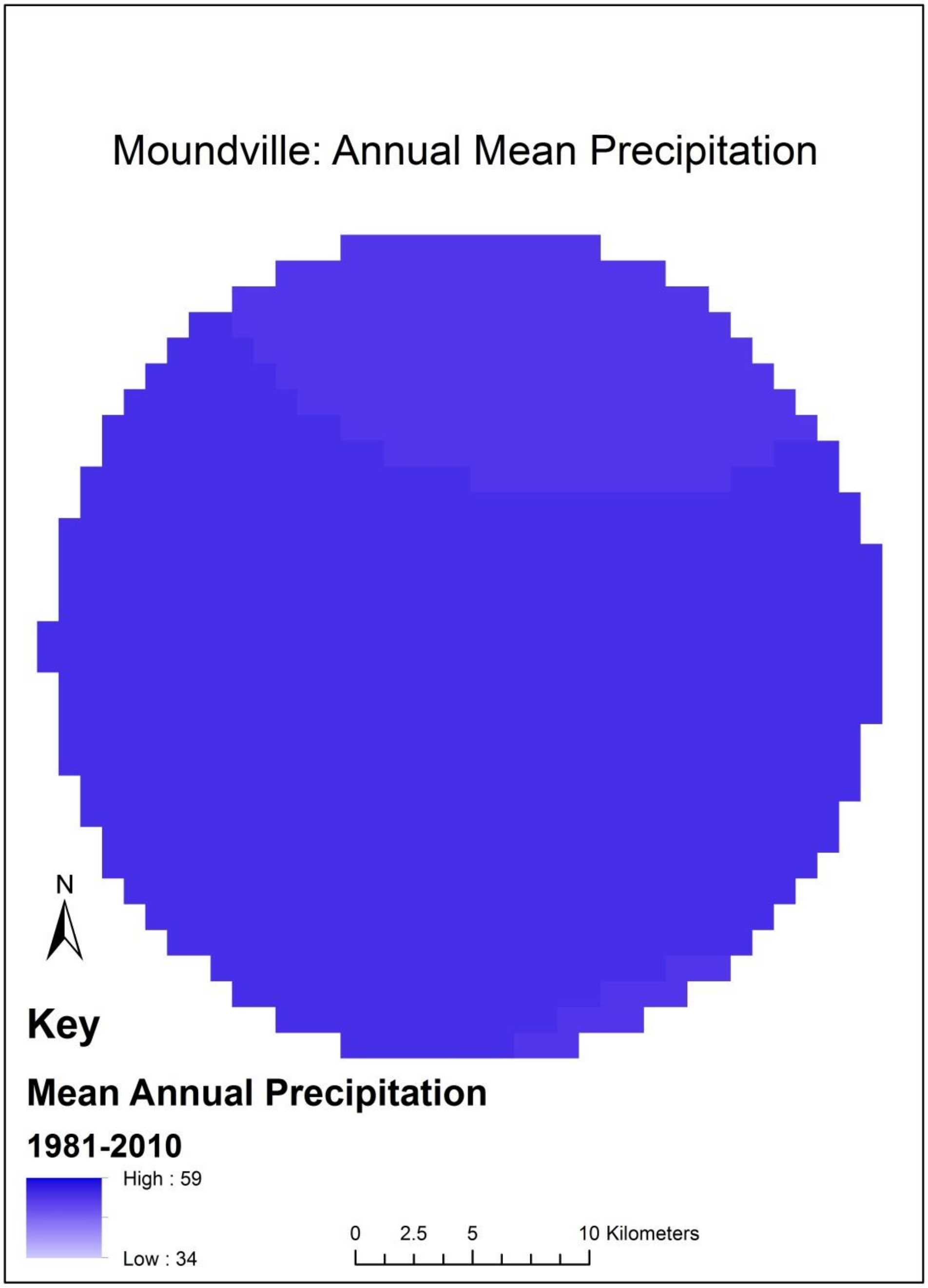

Figure 99. Precipitation at Moundville.

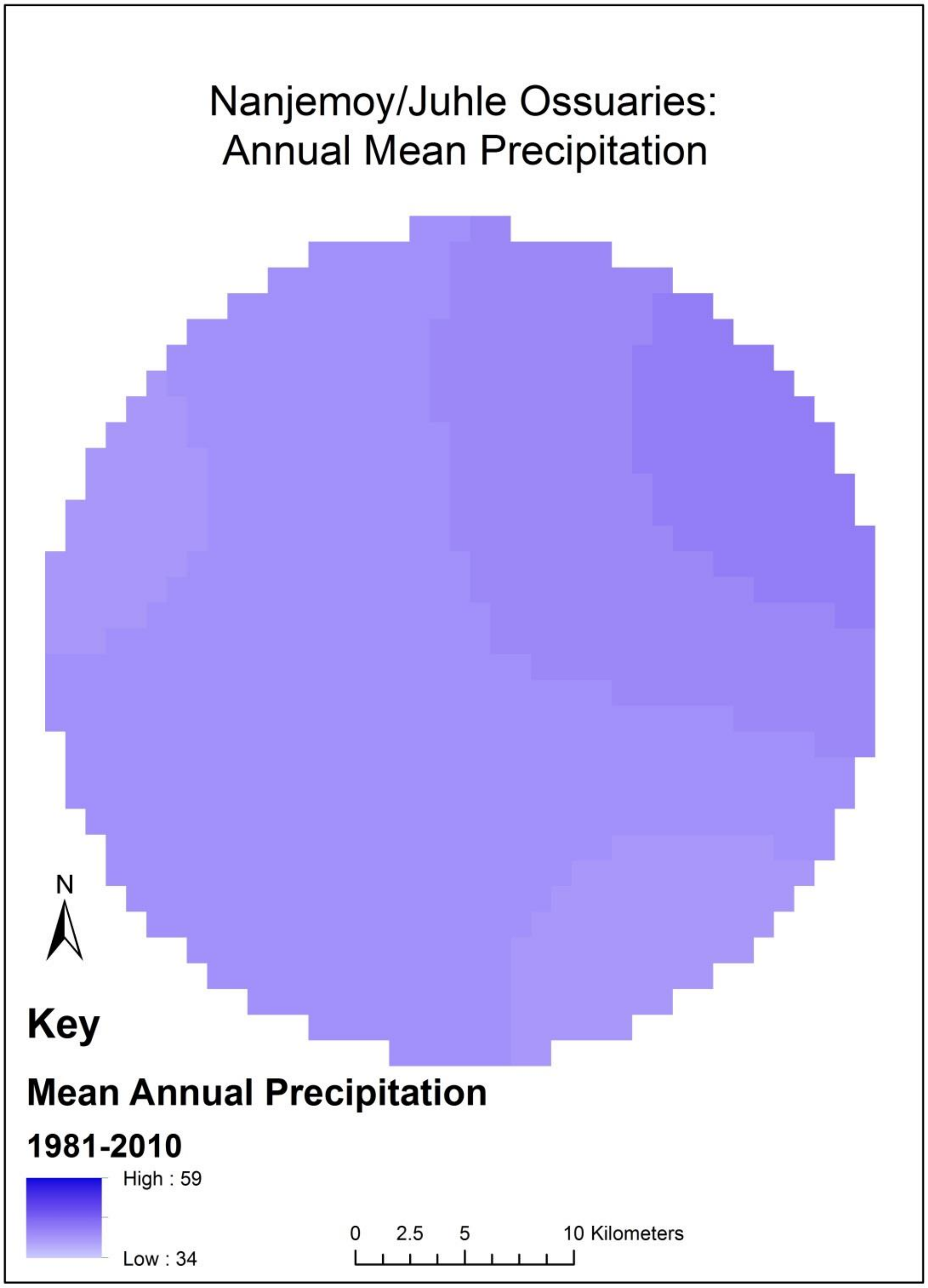

Figure 100. Precipitation at Juhle Ossuary.

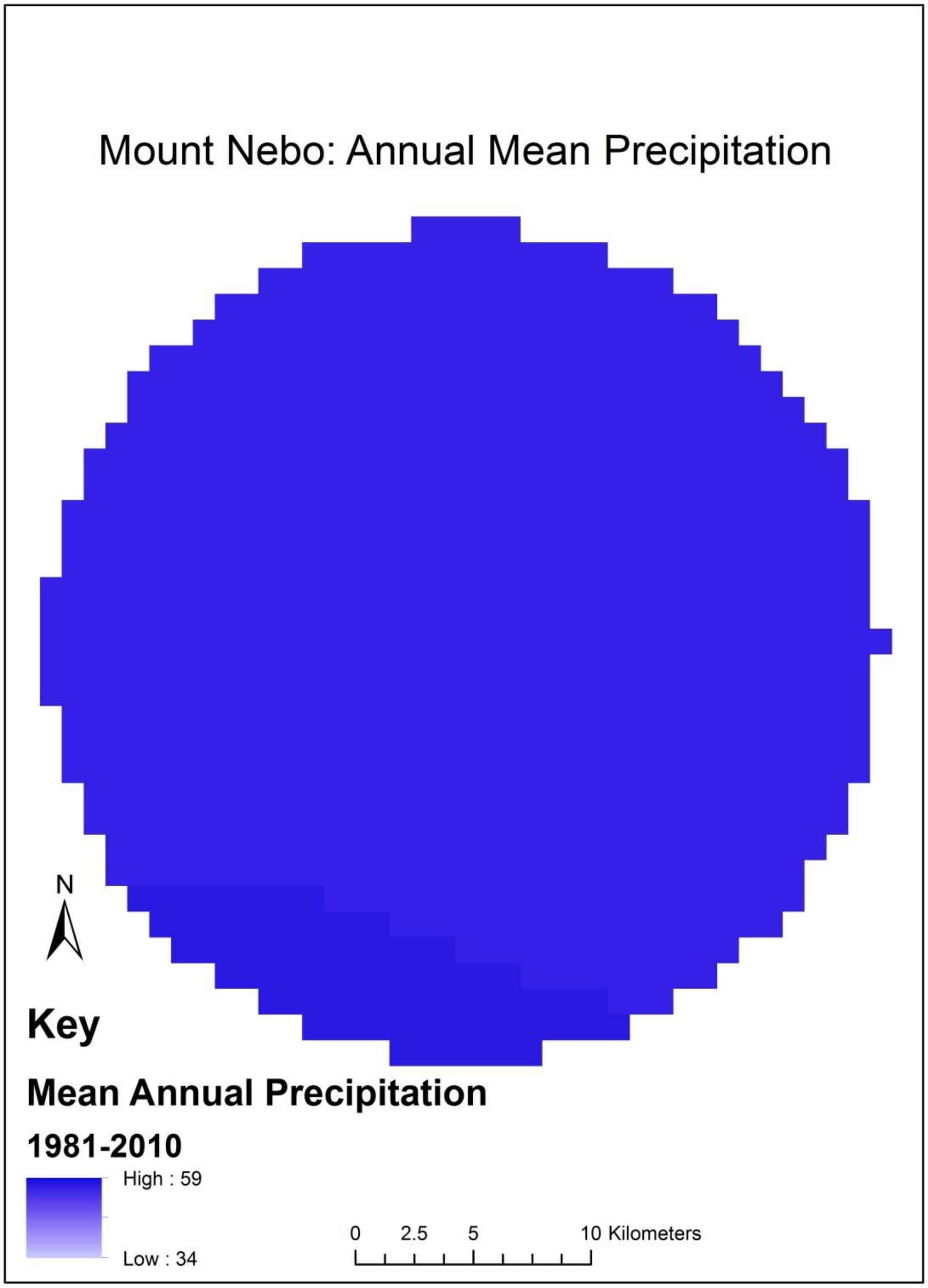

Figure 101. Precipitation at Mount Nebo.

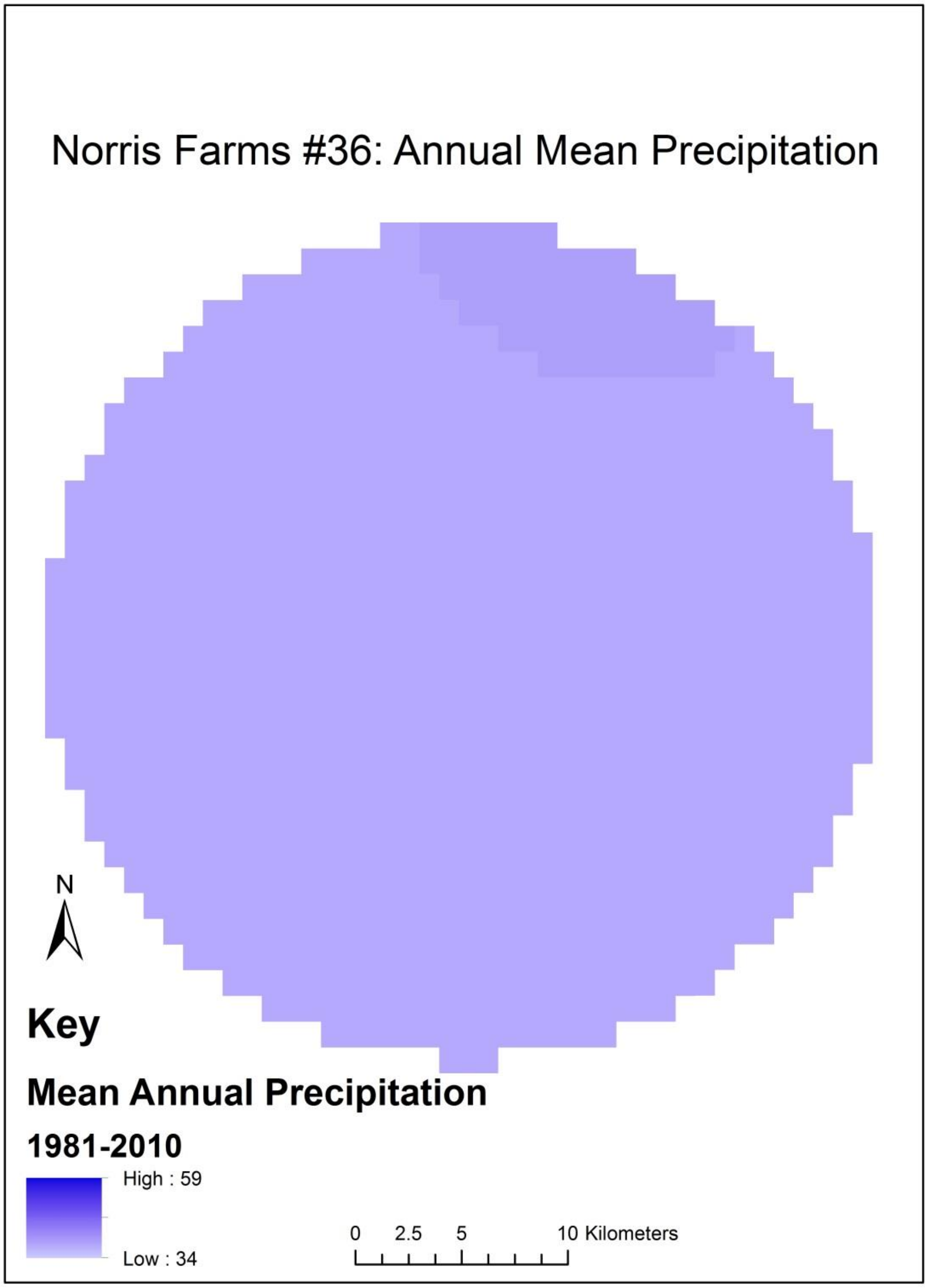

Figure 102. Precipitation at Norris Farms #36.

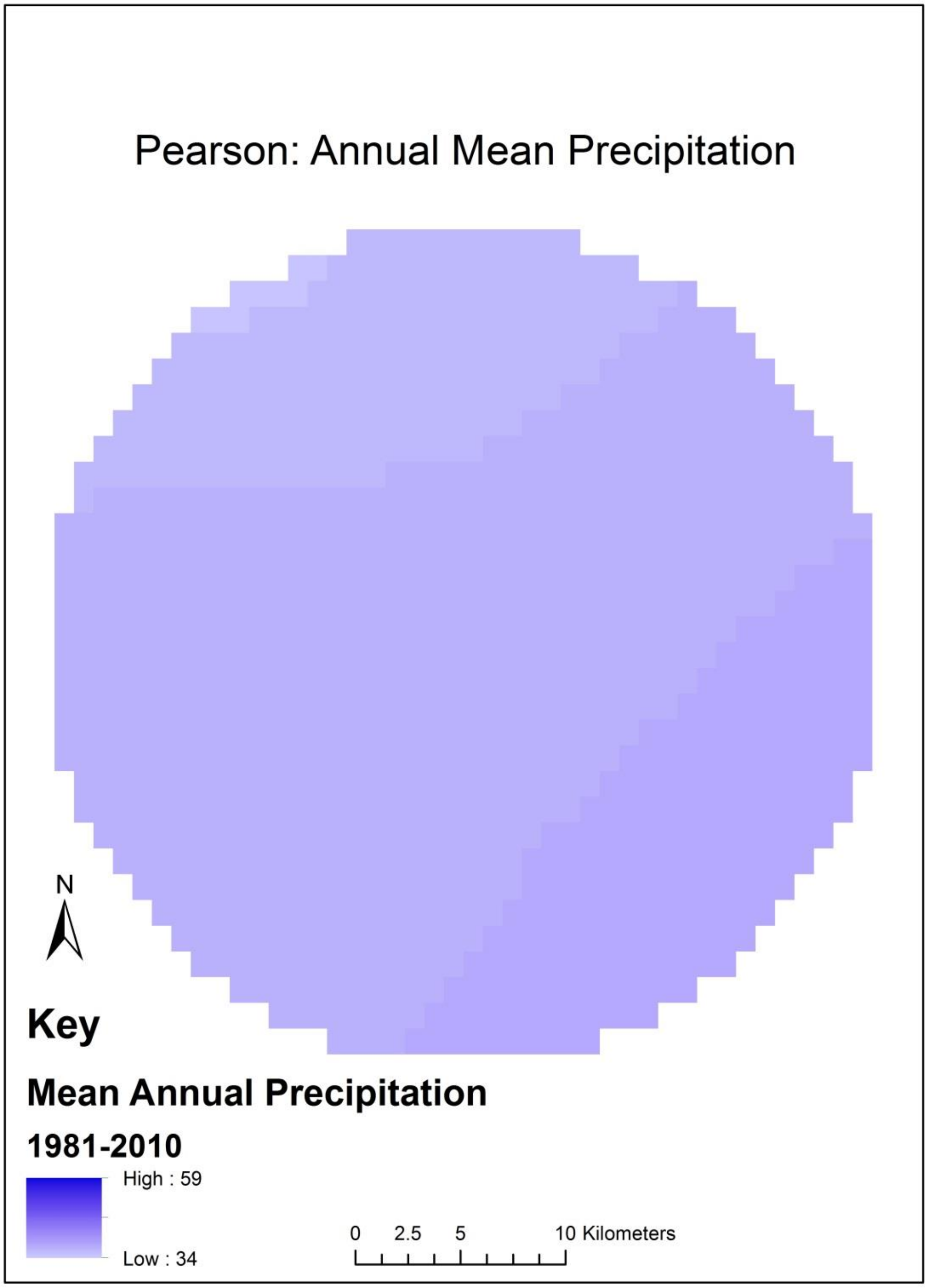

Figure 103. Precipitation at Pearson.

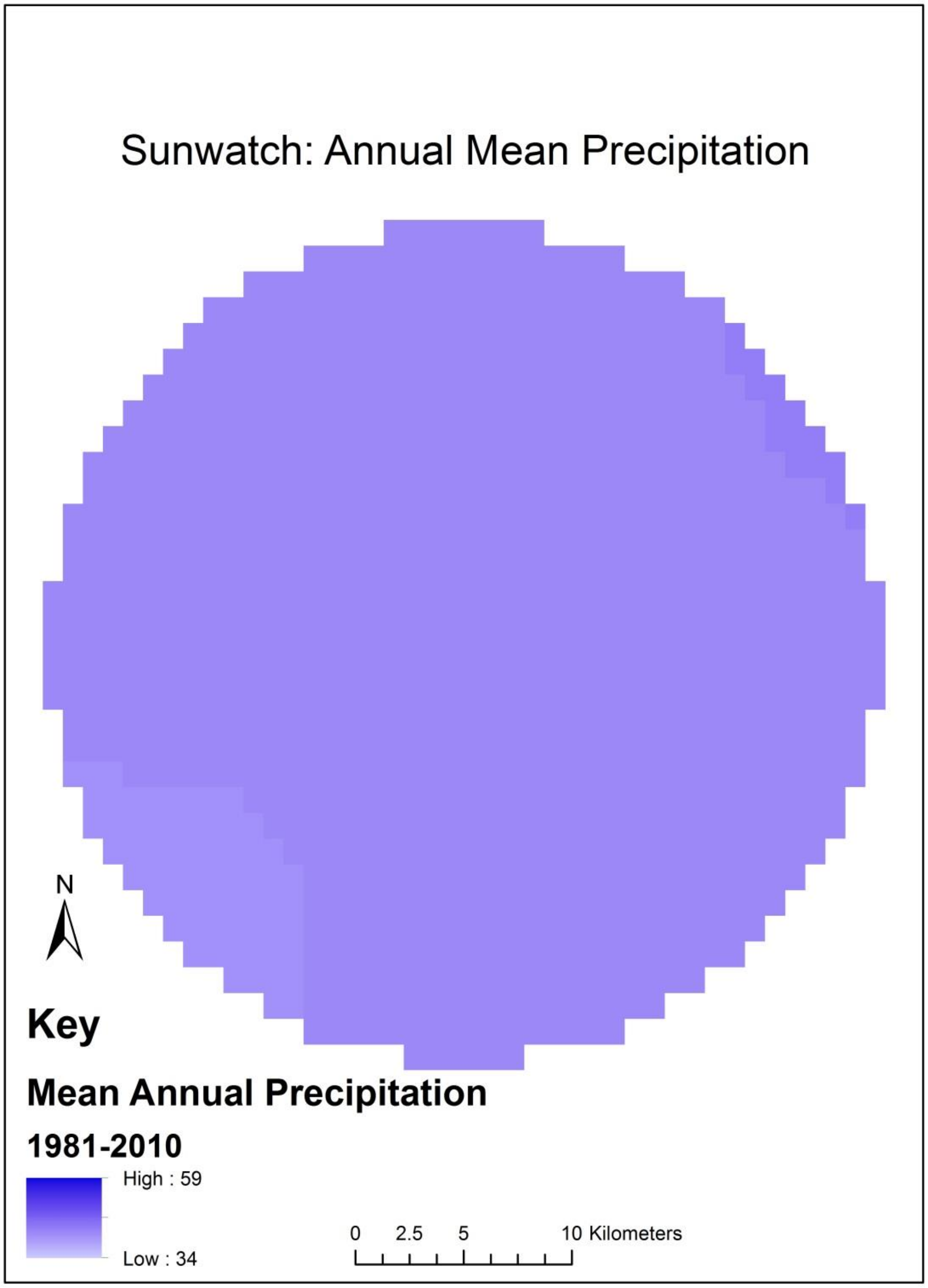

Figure 104. Precipitation at Sunwatch.

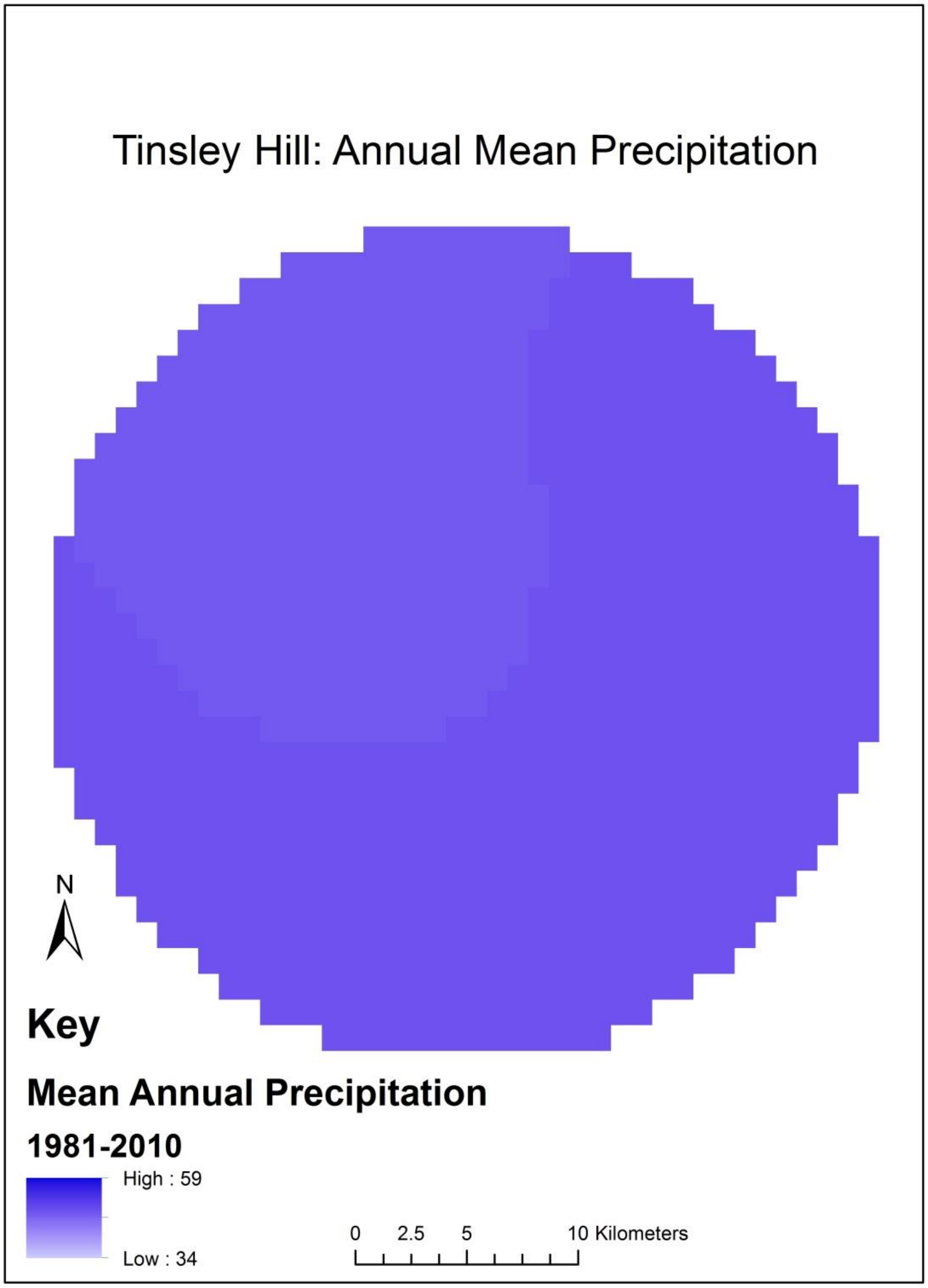

Figure 105. Precipitation at Tinsley Hill.

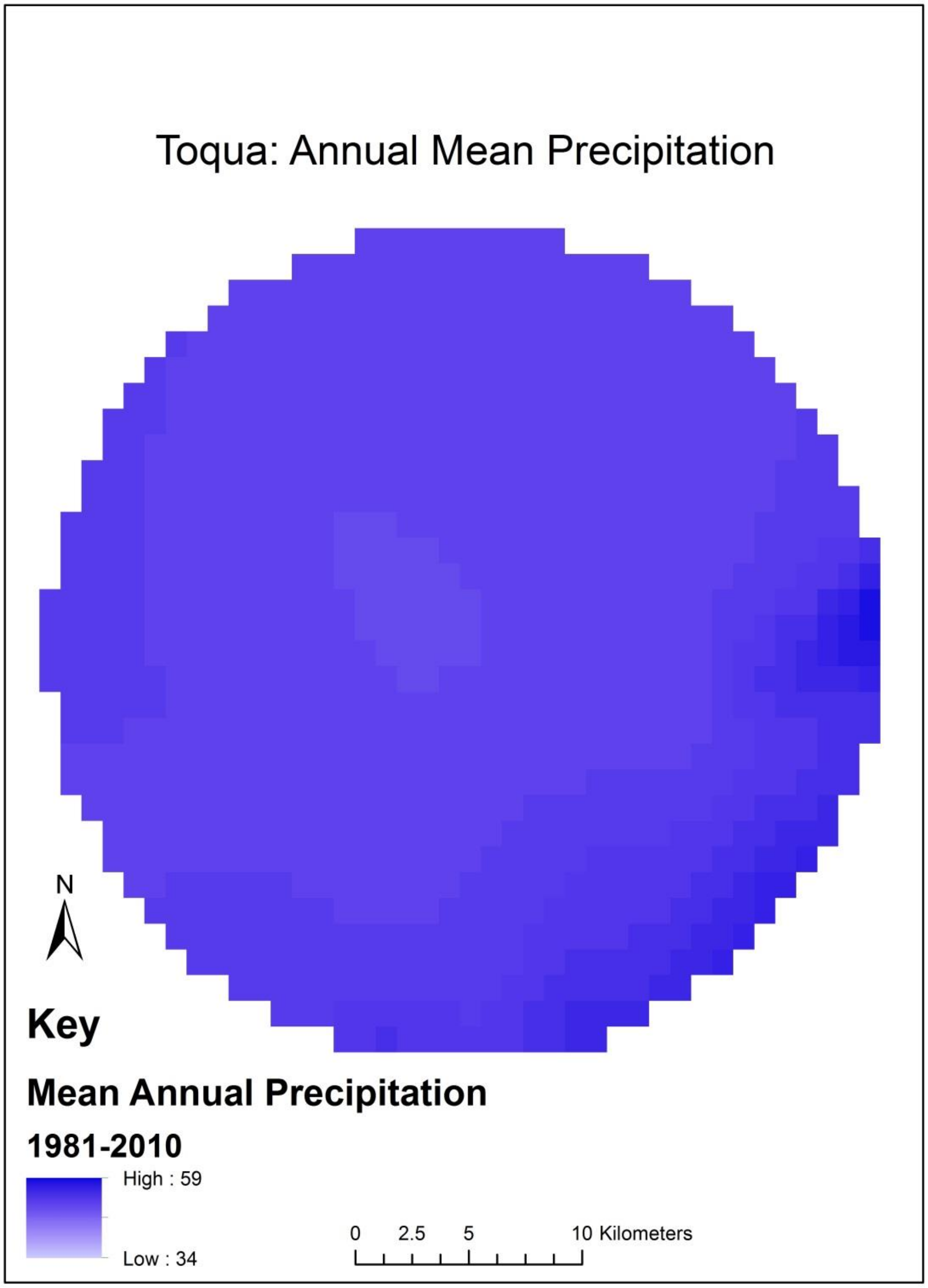

Figure 106. Precipitation at Toqua.

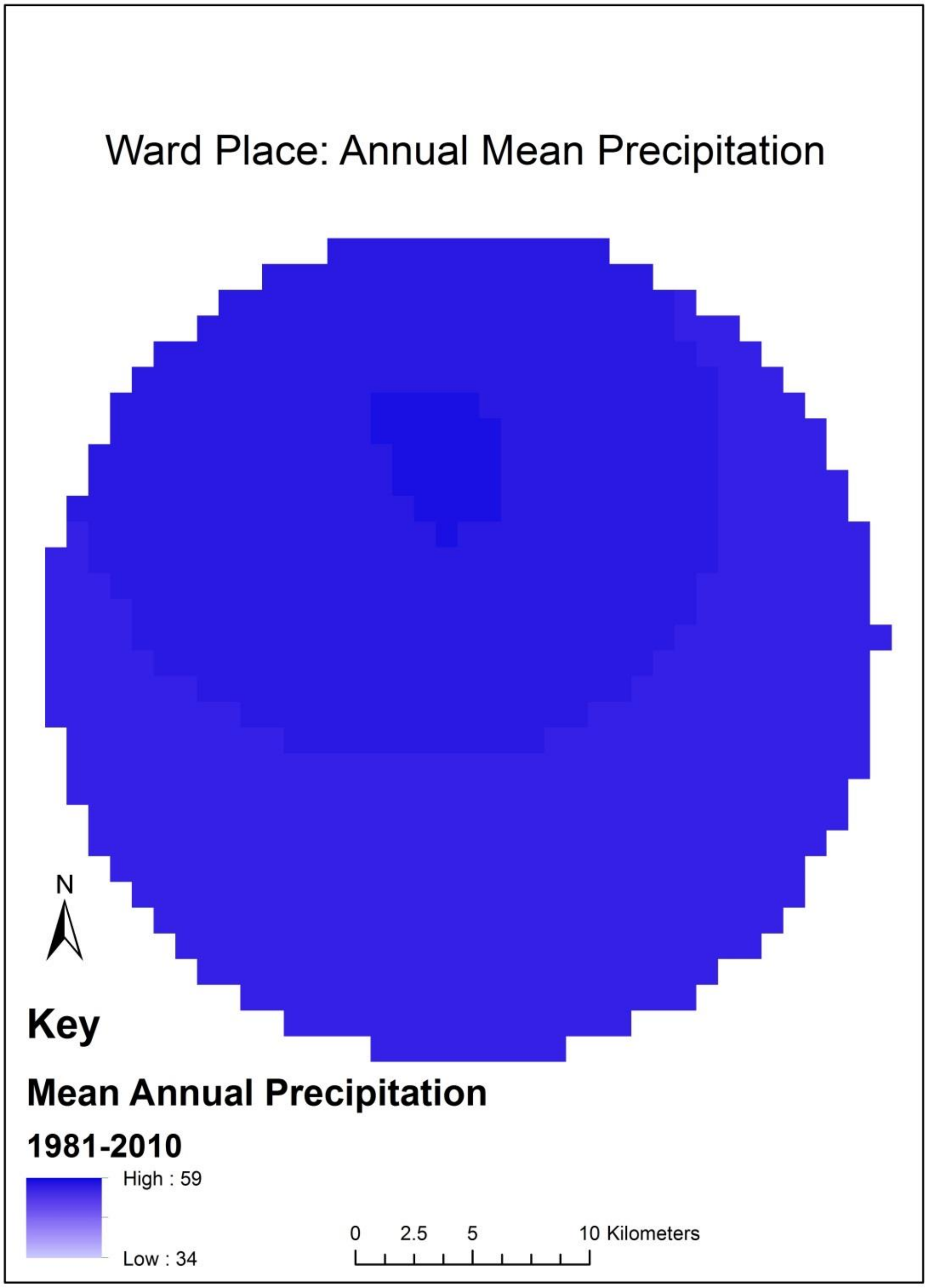

Figure 107. Precipitation at Ward Place.

A5. Surface Area of Major Lakes and Rivers

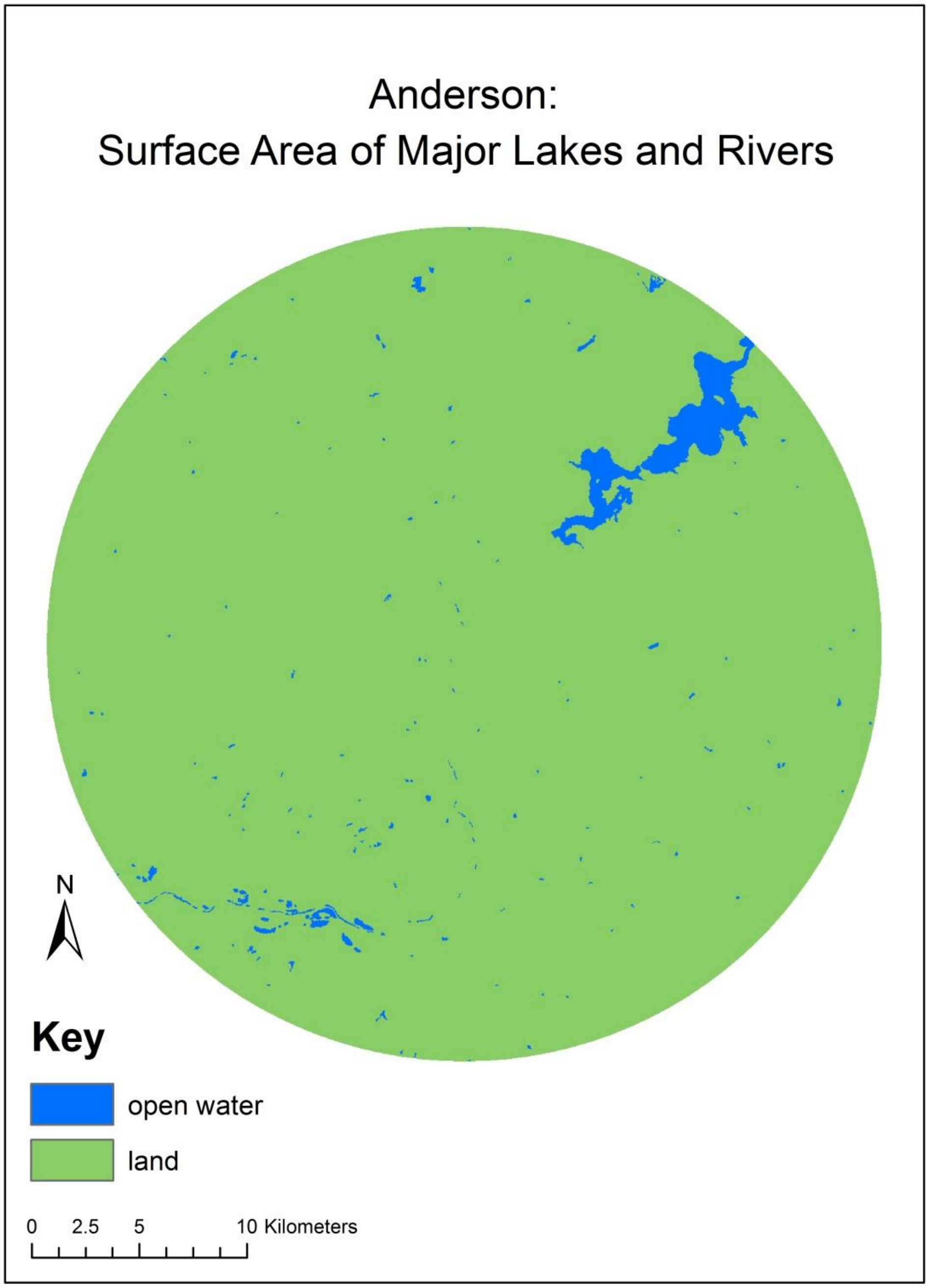

Figure 108. Lakes and rivers at Anderson.

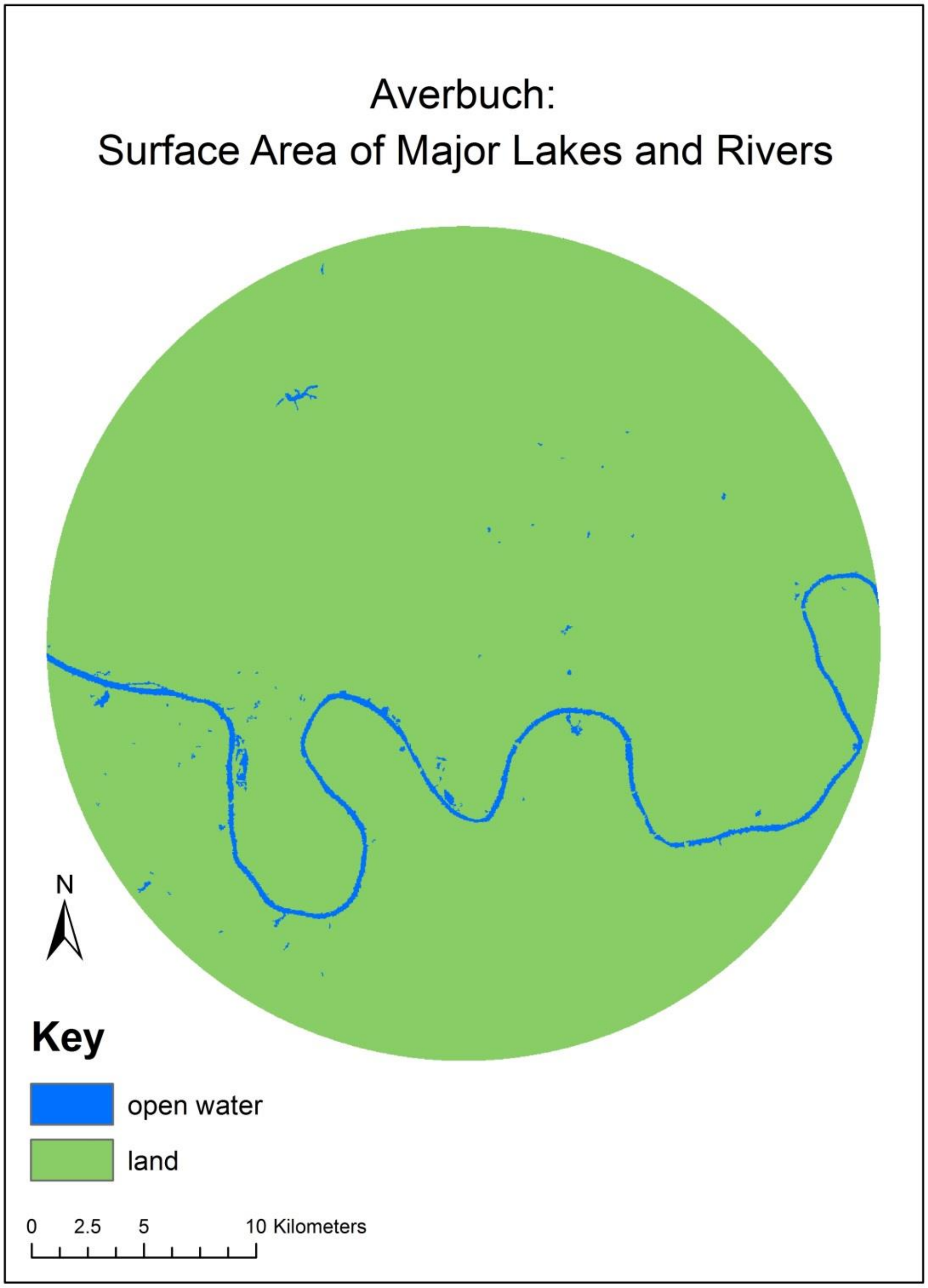

Figure 109. Lakes and rivers at Averbuch.

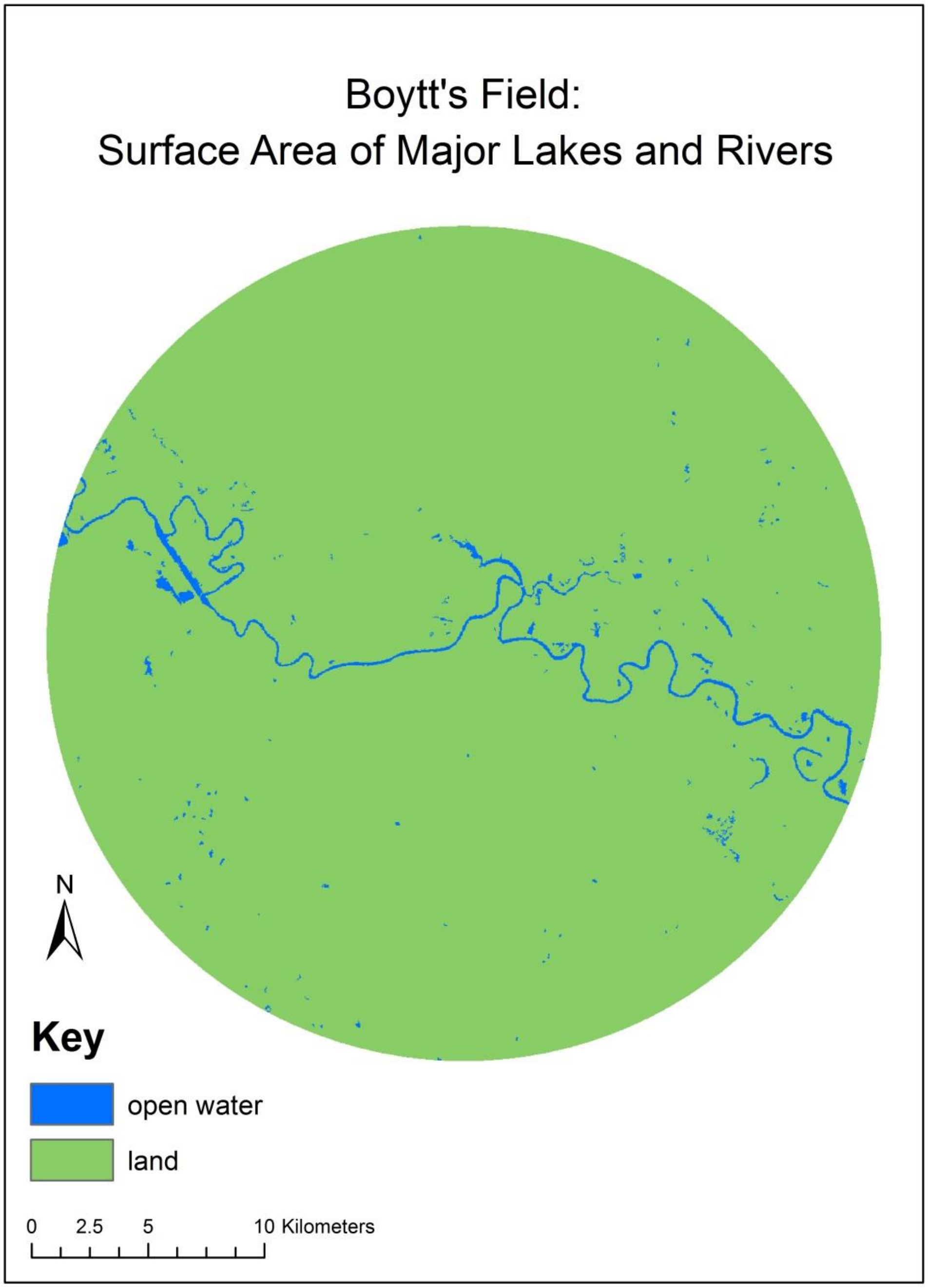

Figure 110. Lakes and rivers at Boytt's Field.

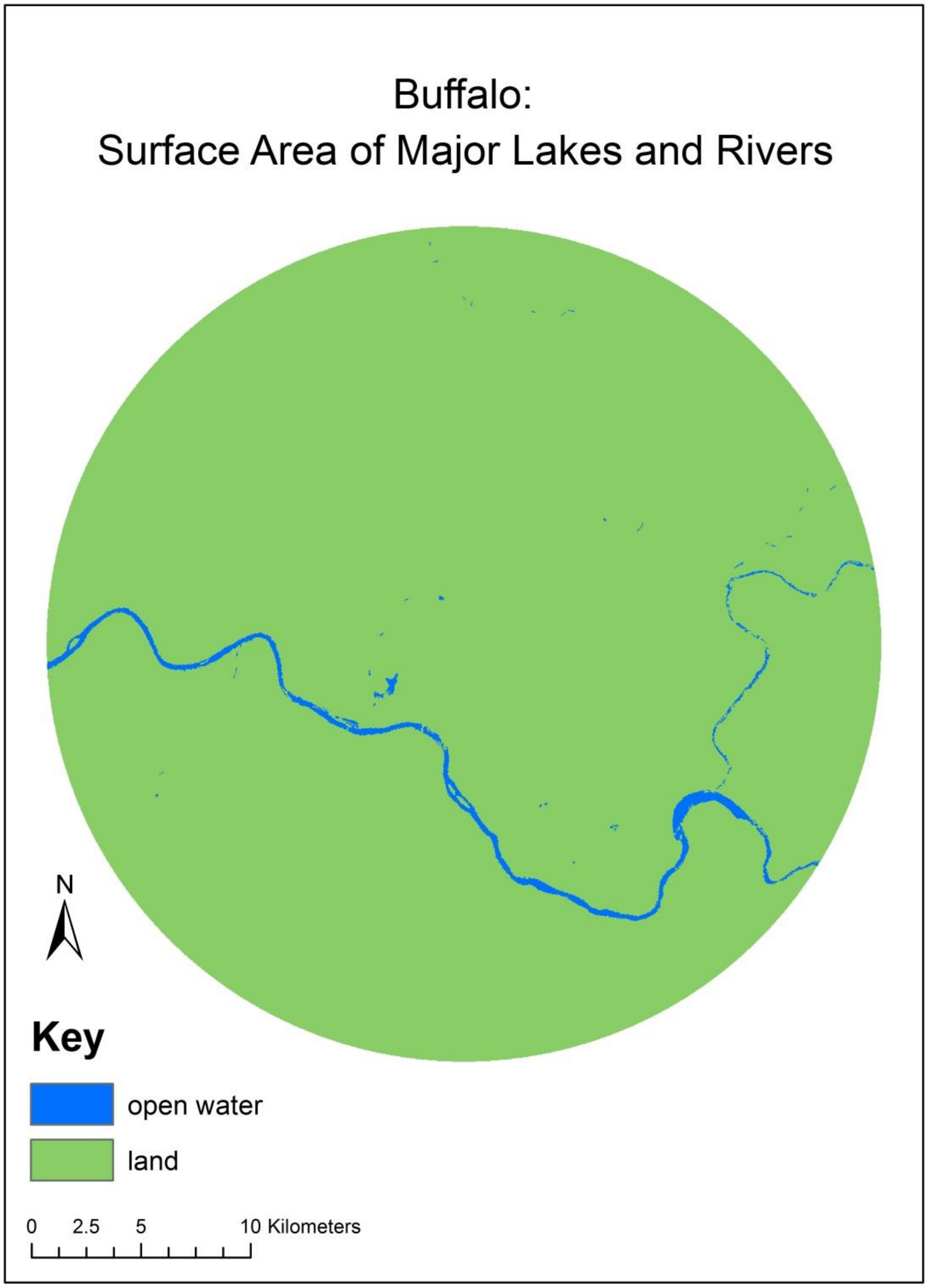

Figure 111. Lakes and rivers at Buffalo.

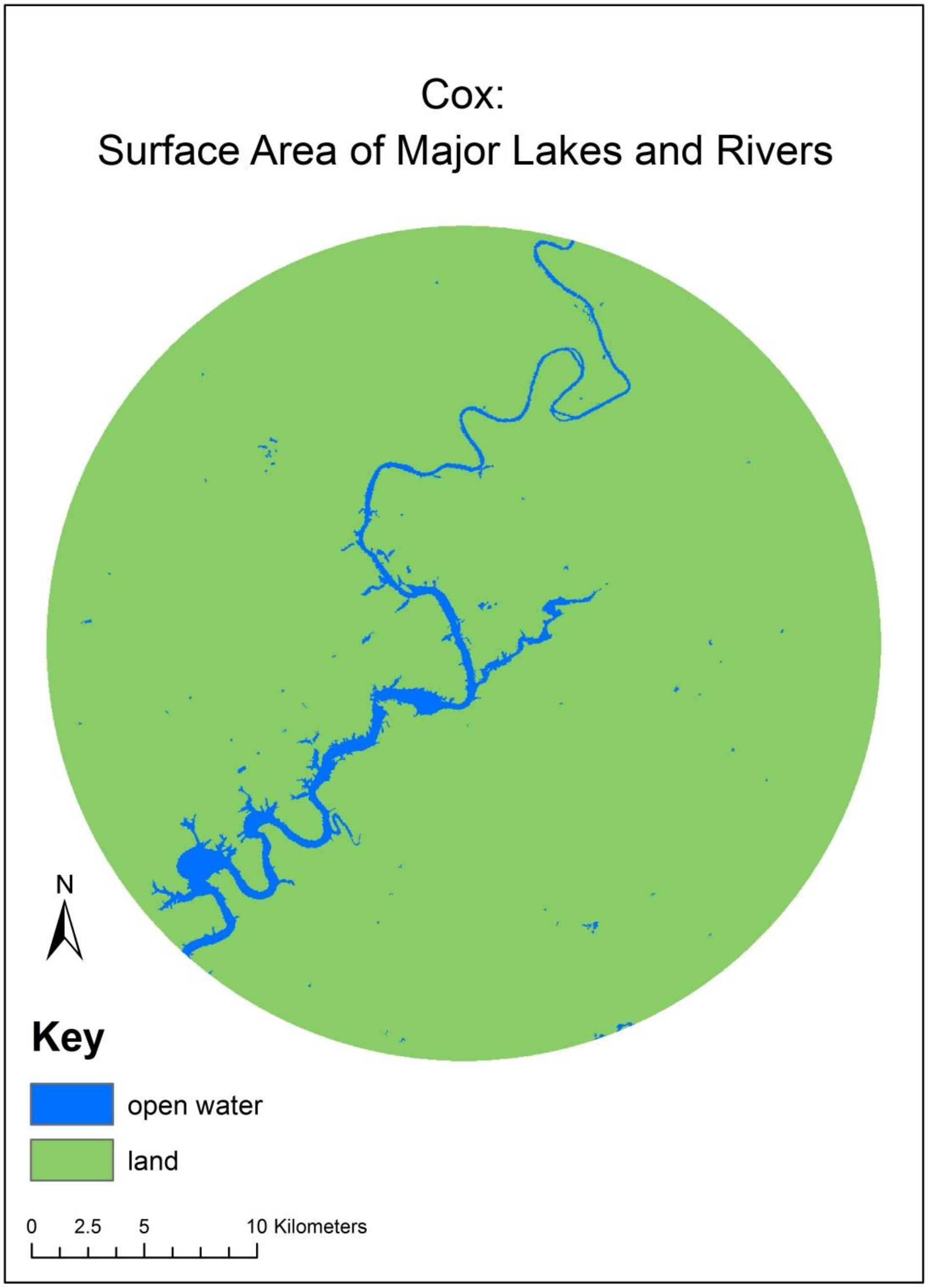

Figure 112. Lakes and rivers at Cox.

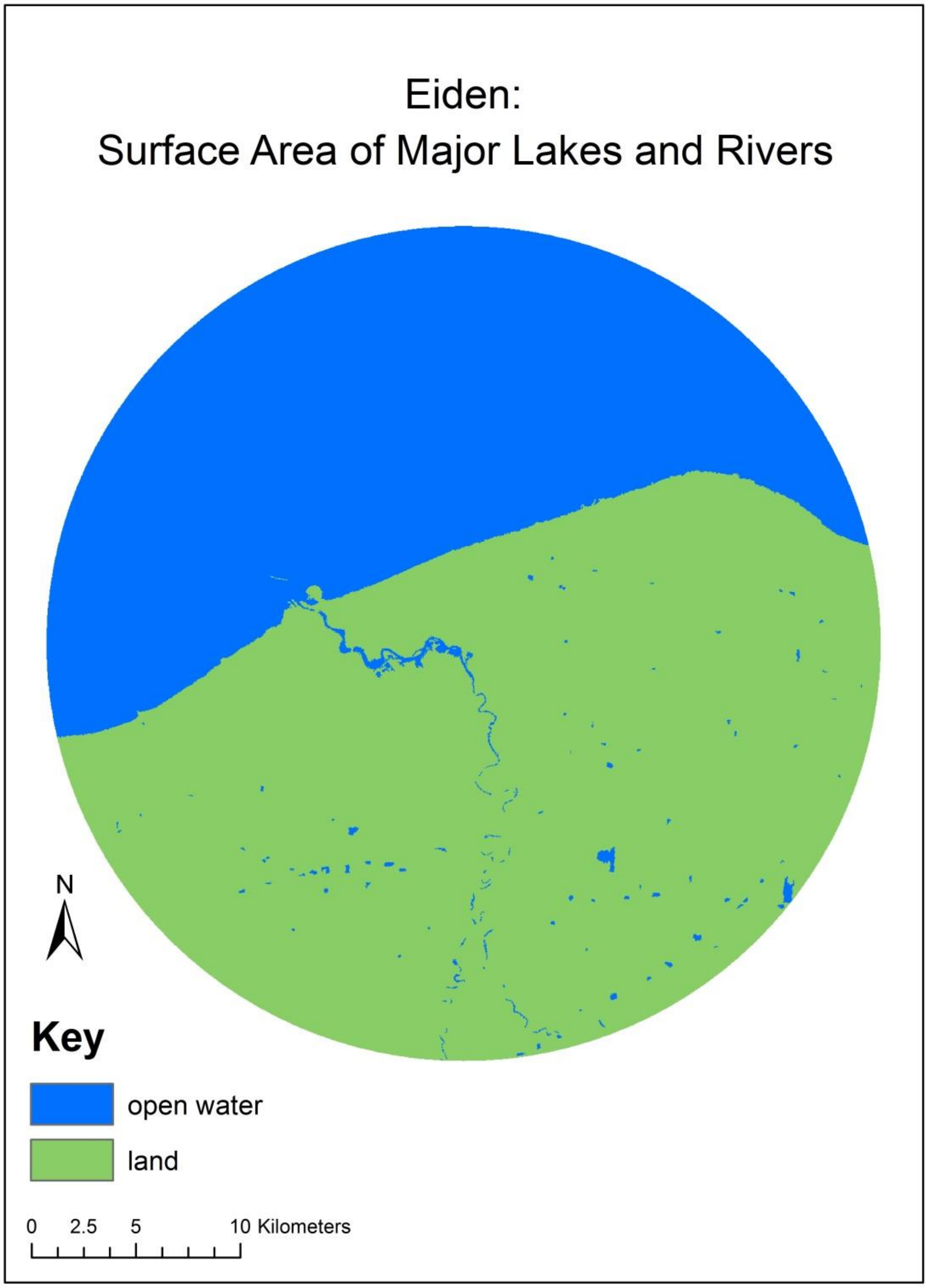

Figure 113. Lakes and rivers at Eiden.

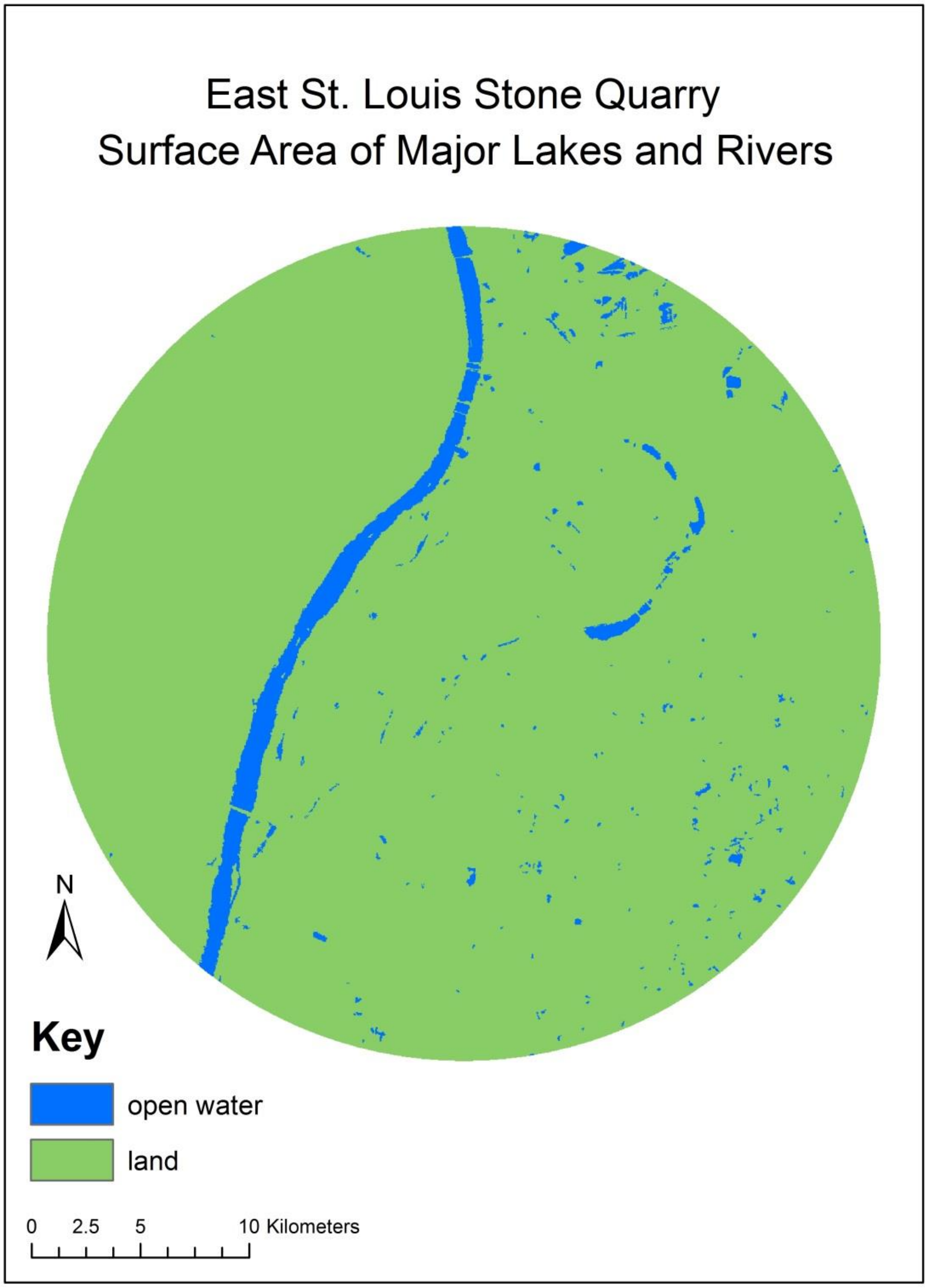

Figure 114. Lakes and rivers at East St. Louis Stone Quarry.

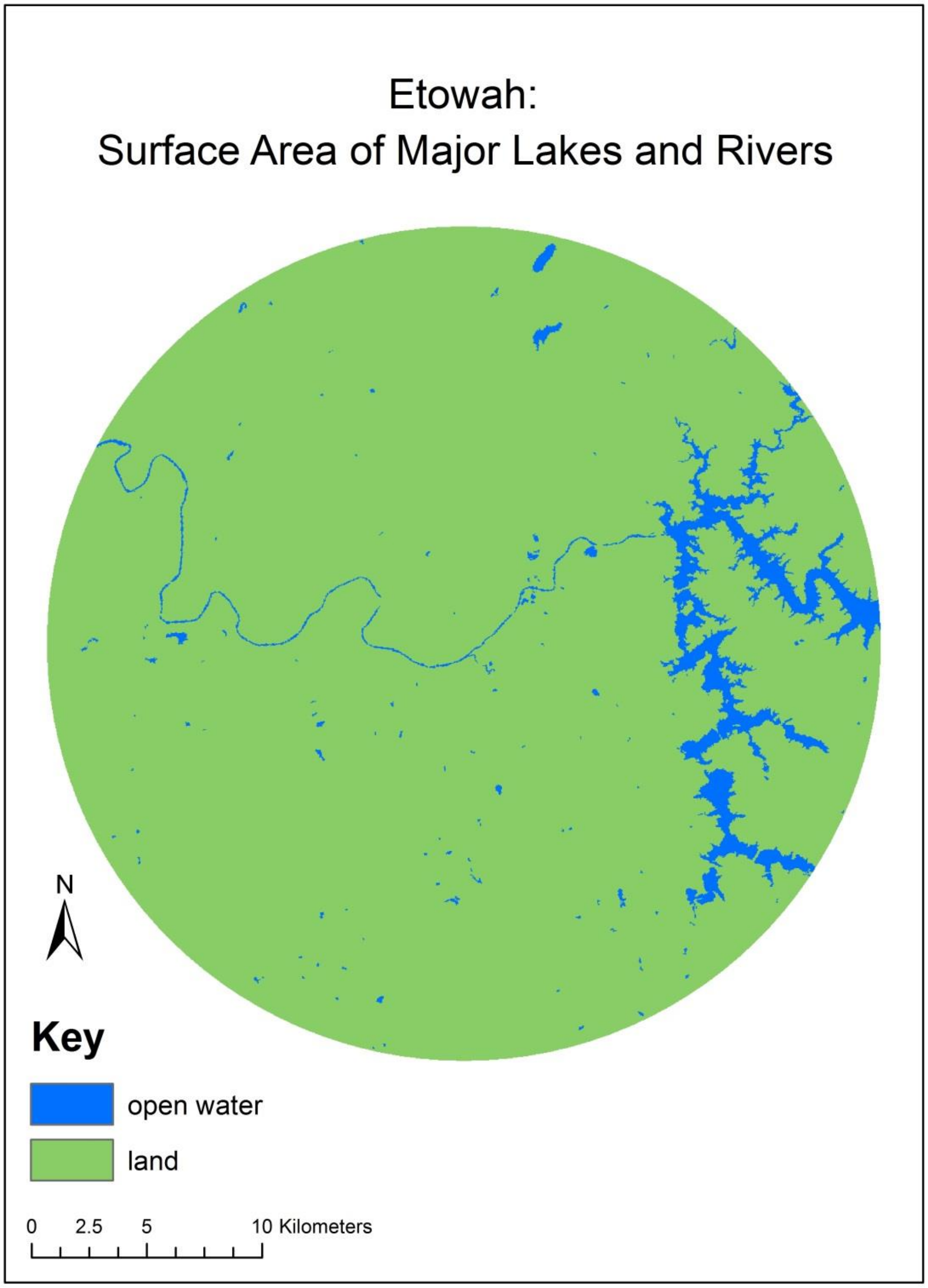

Figure 115. Lakes and rivers at Etowah.

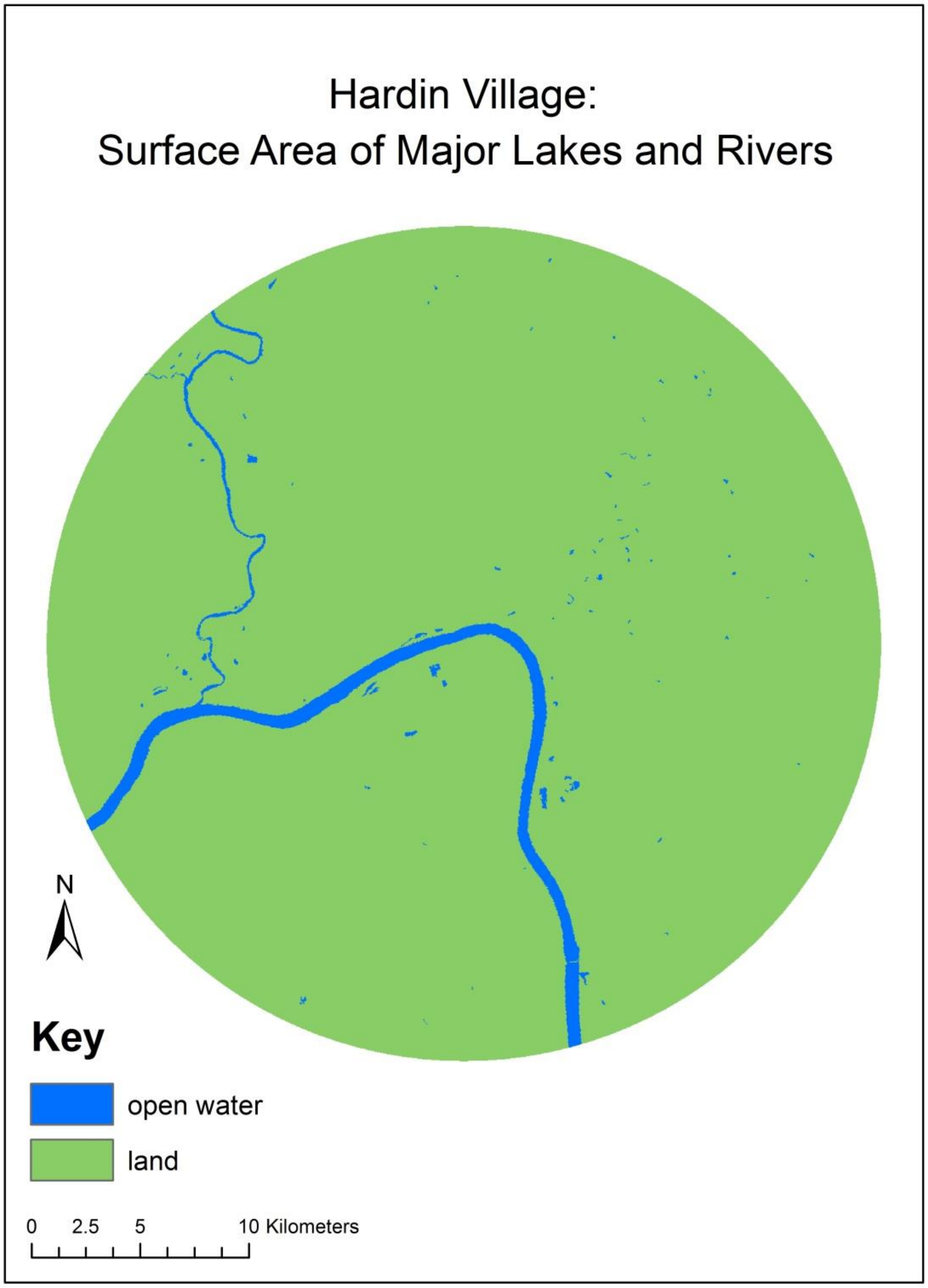

Figure 116. Lakes and rivers at Hardin Village.

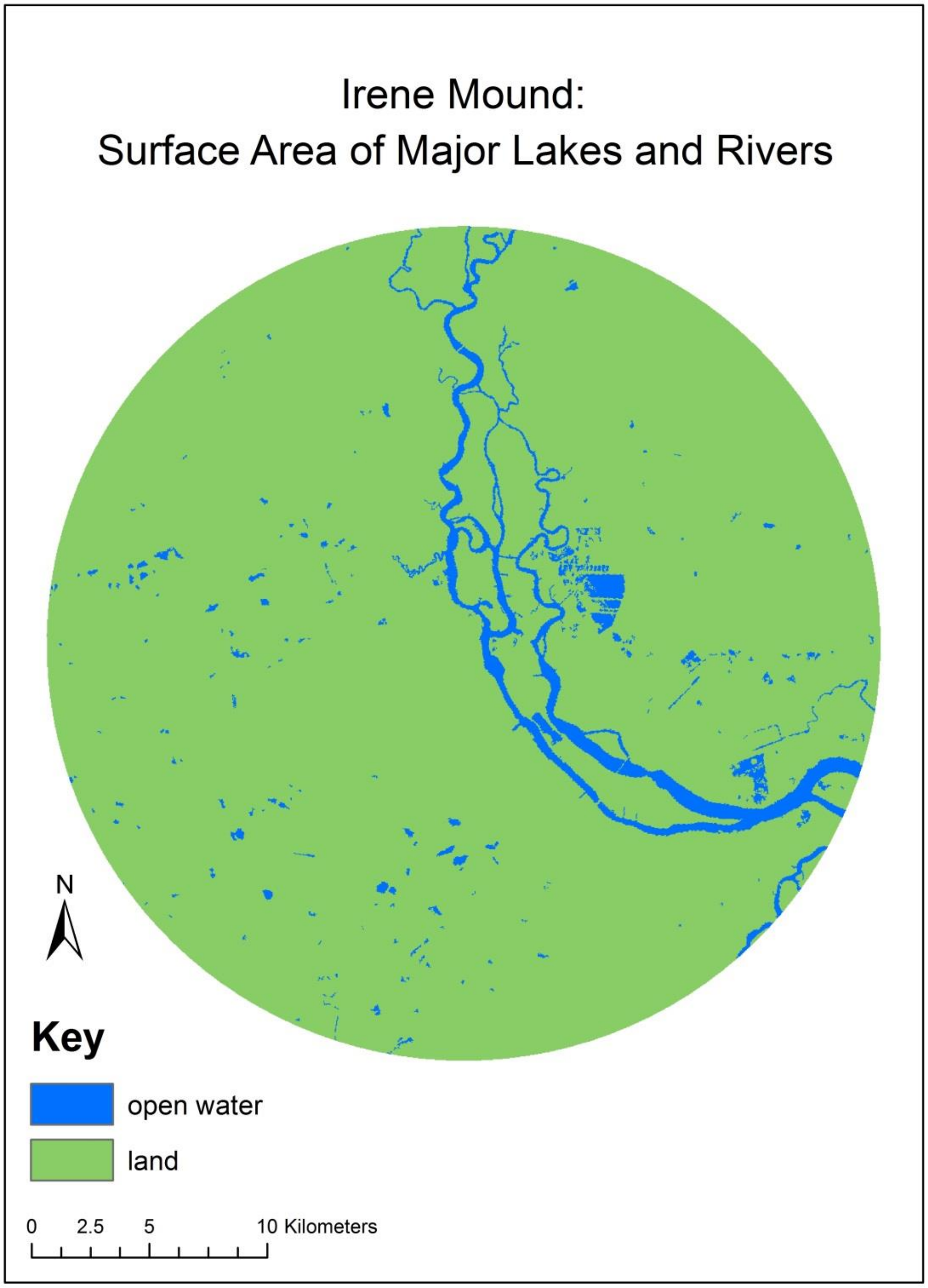

Figure 117. Lakes and rivers at Irene Mound.

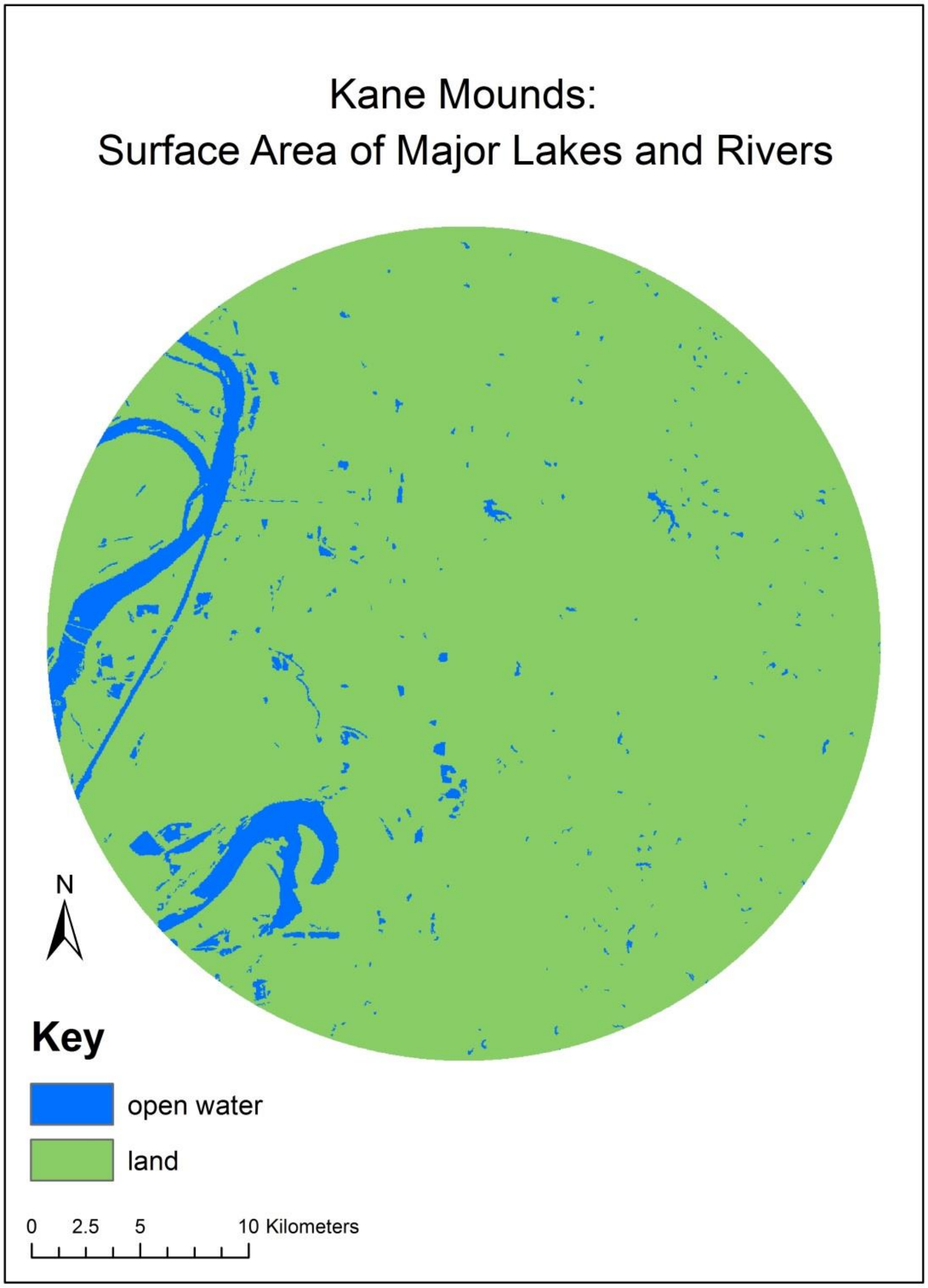

Figure 118. Lakes and rivers at Kane Mounds.

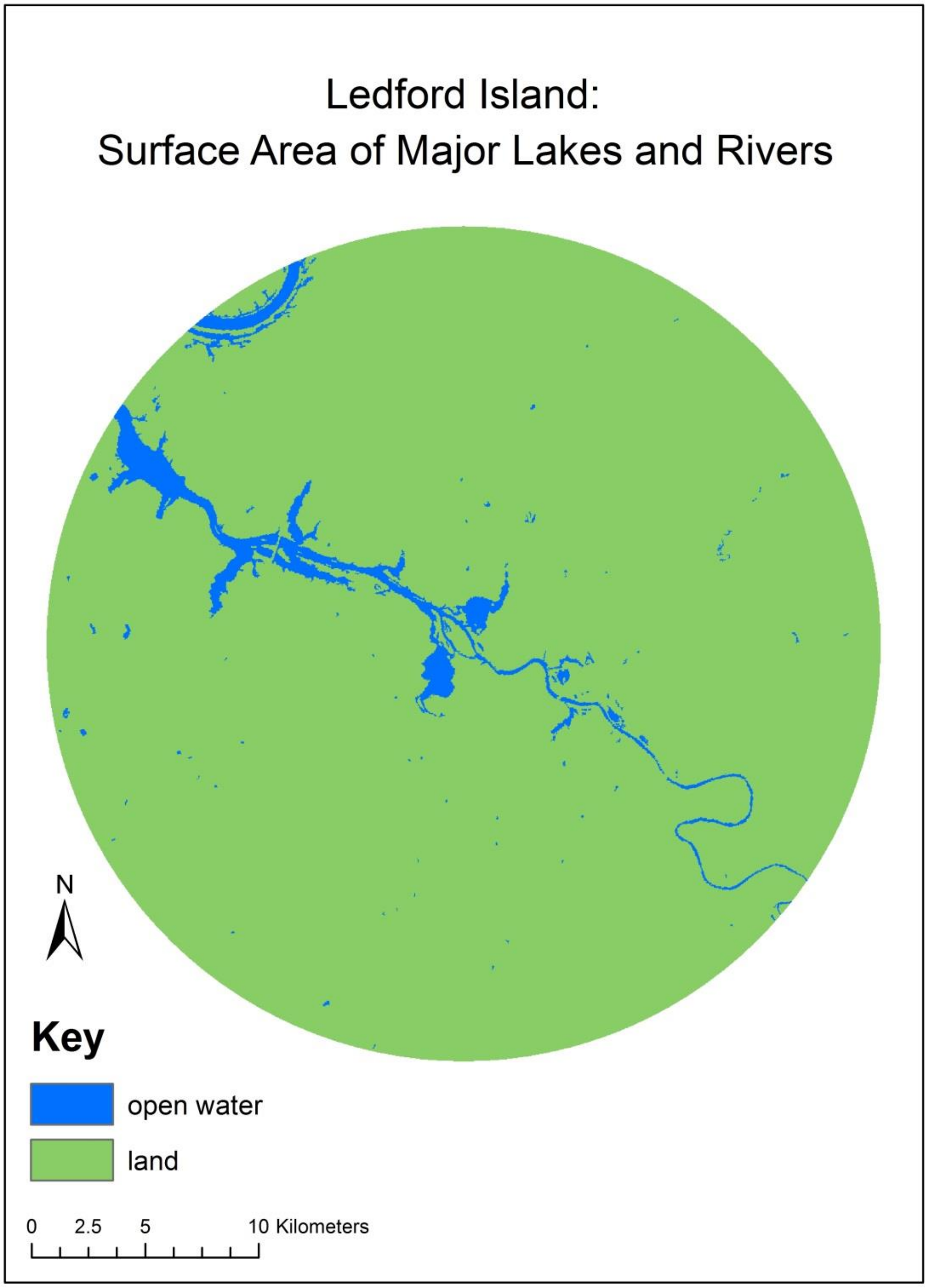

Figure 119. Lakes and rivers at Ledford Island.

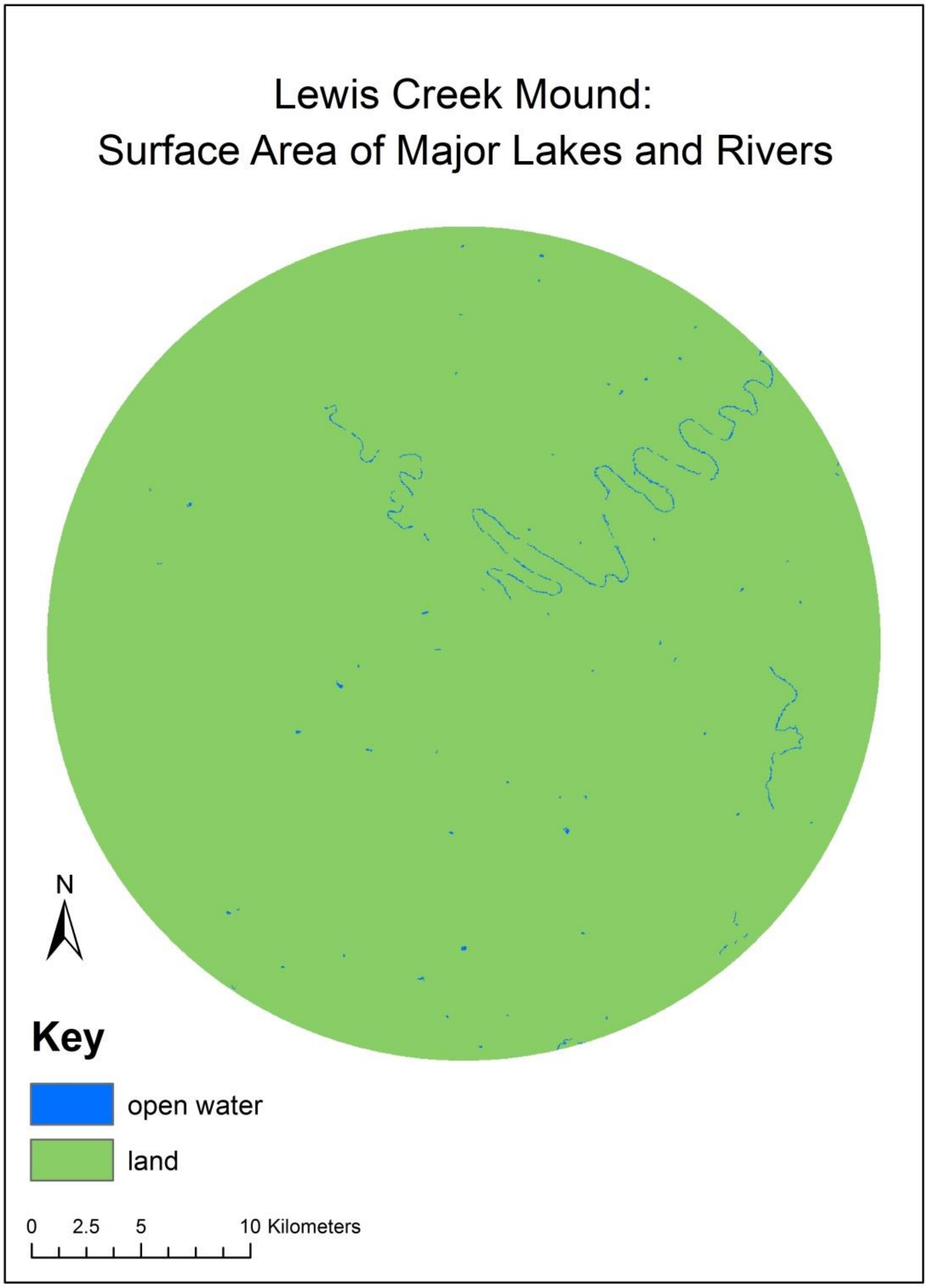

Figure 120. Lakes and rivers at Lewis Creek Mound.

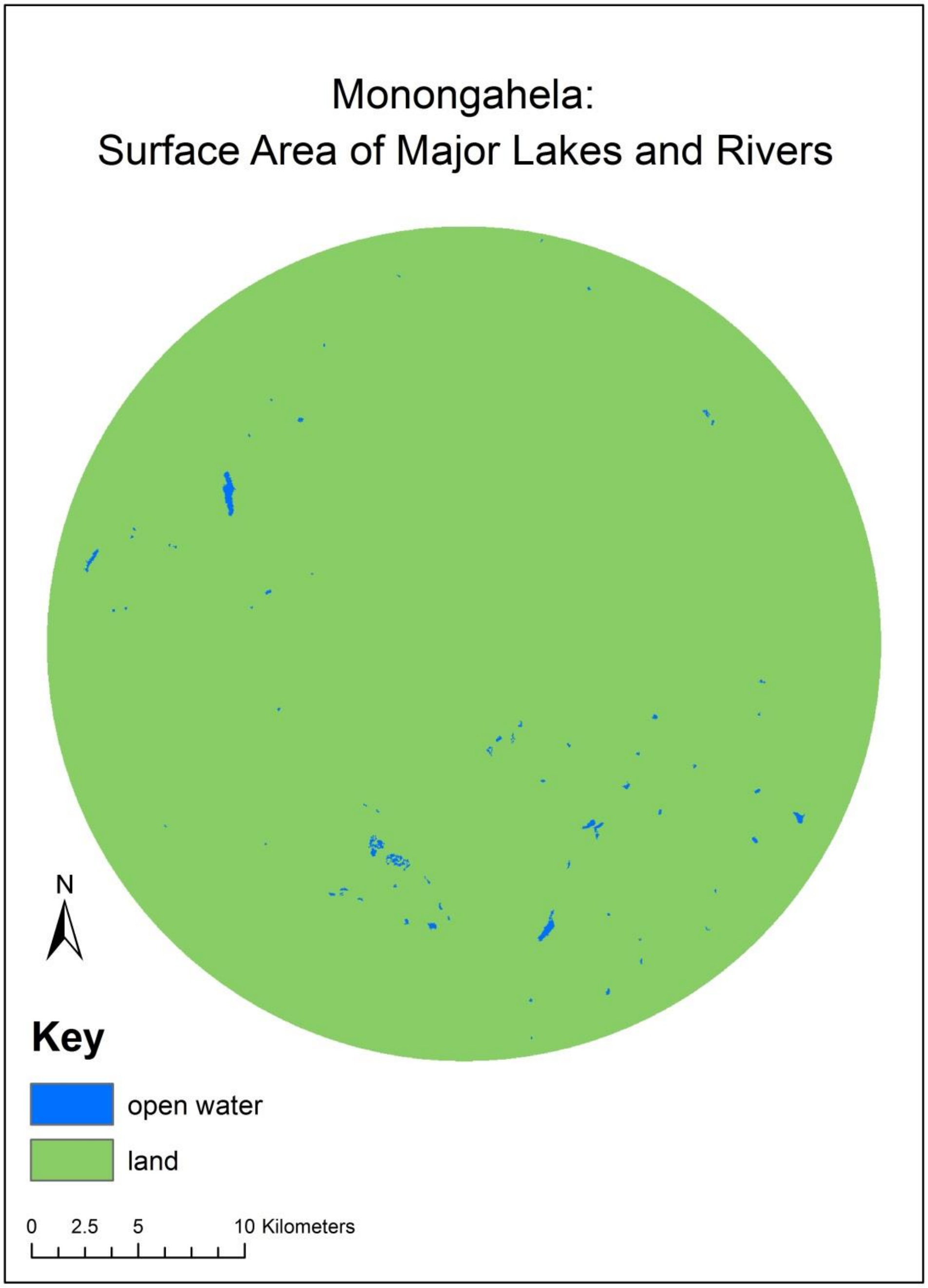

Figure 121. Lakes and rivers at Monongahela.

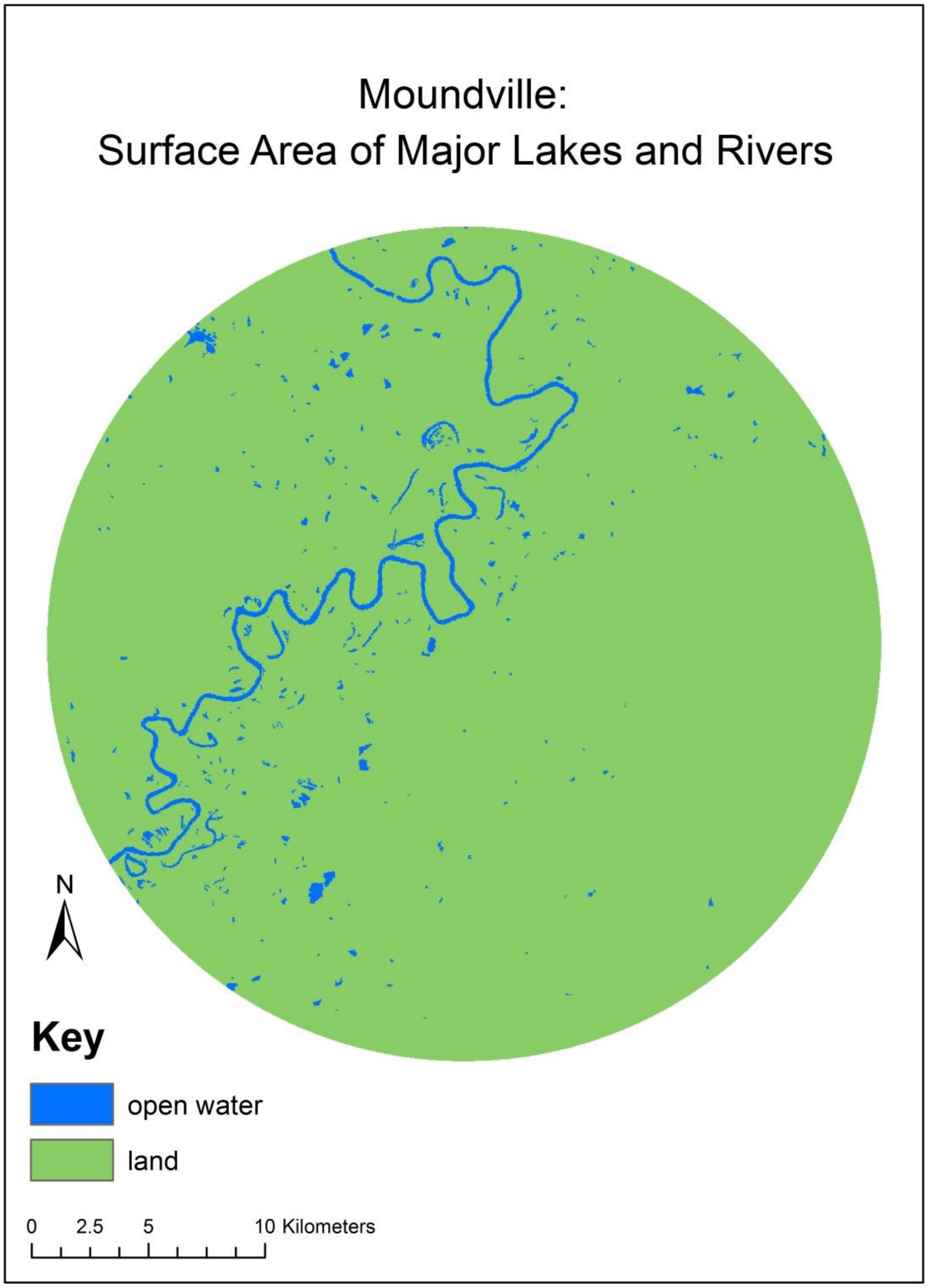

Figure 122. Lakes and rivers at Moundville.

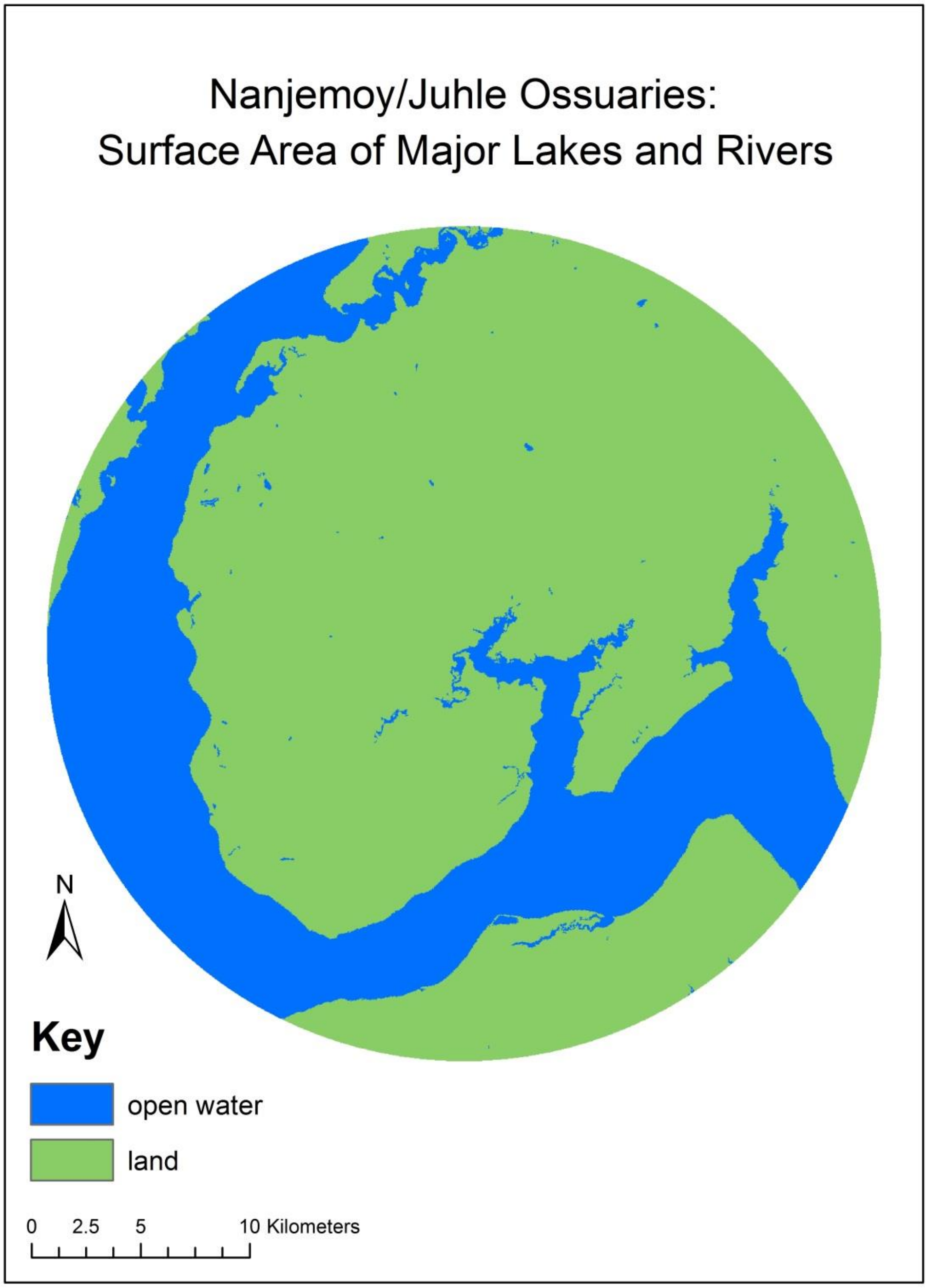

Figure 123. Lakes and rivers at Juhle Ossuary.

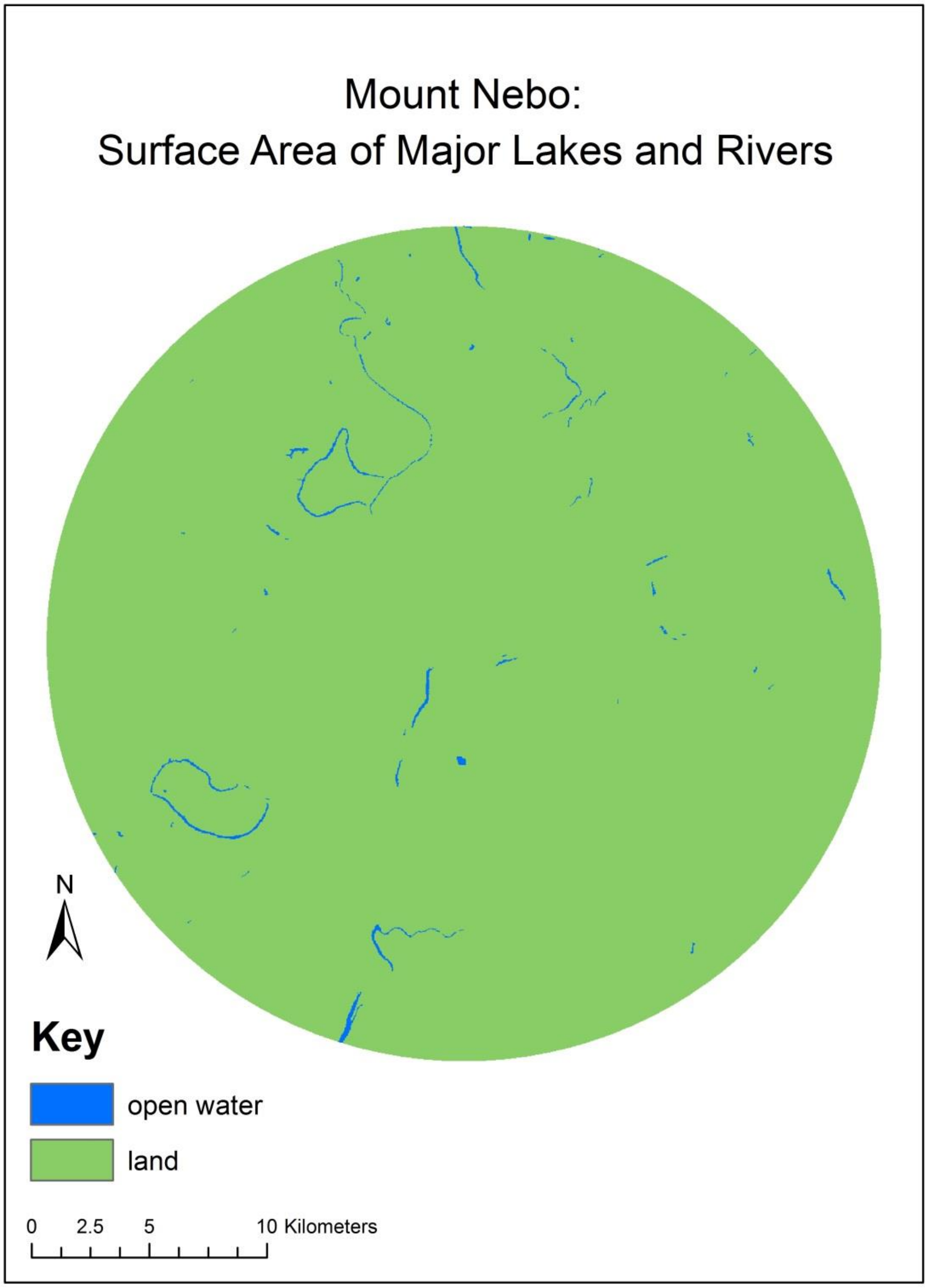

Figure 124. Lakes and rivers at Mount Nebo.

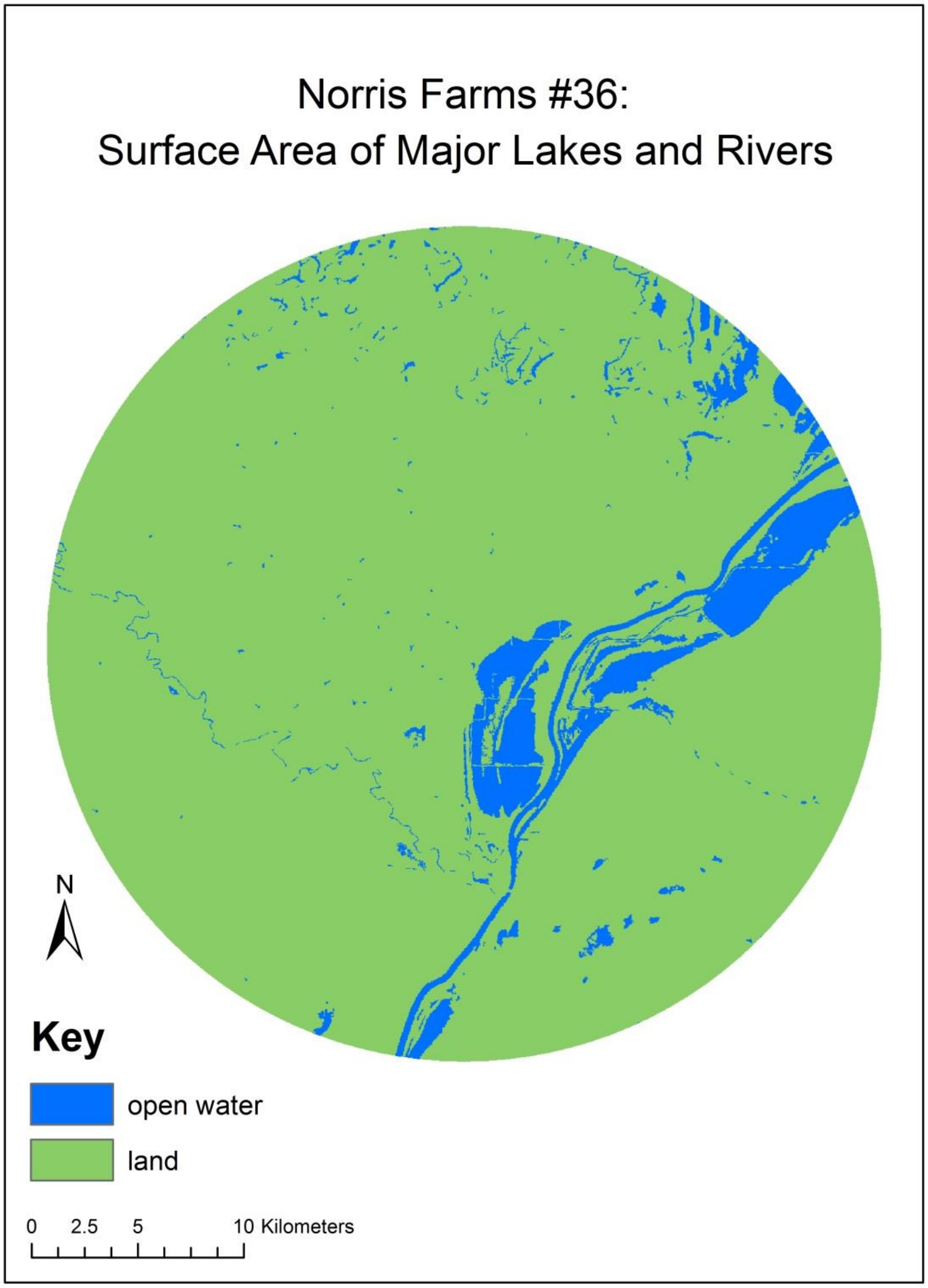

Figure 125. Lakes and rivers at Norris Farms #36.

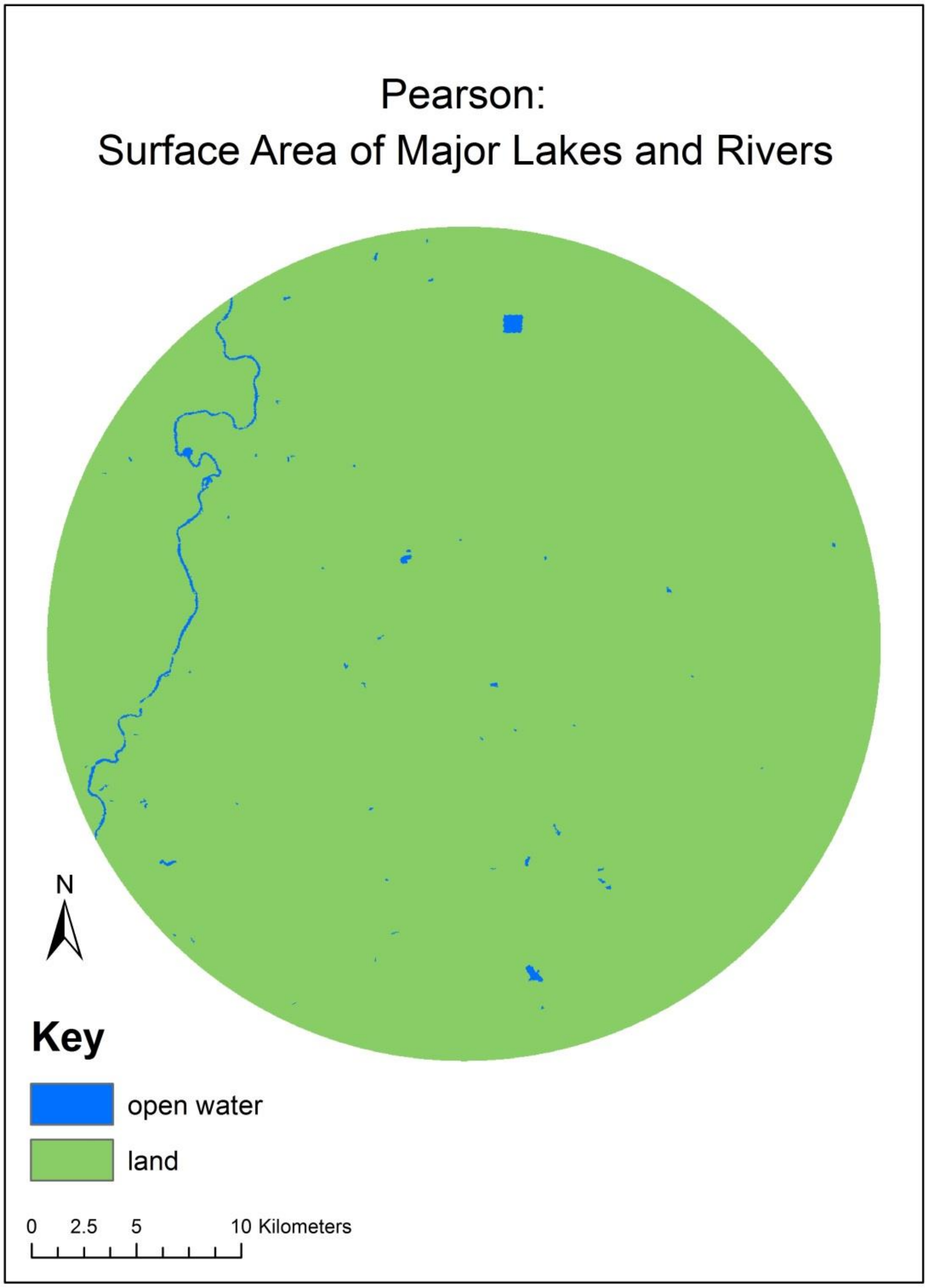

Figure 126. Lakes and rivers at Pearson.

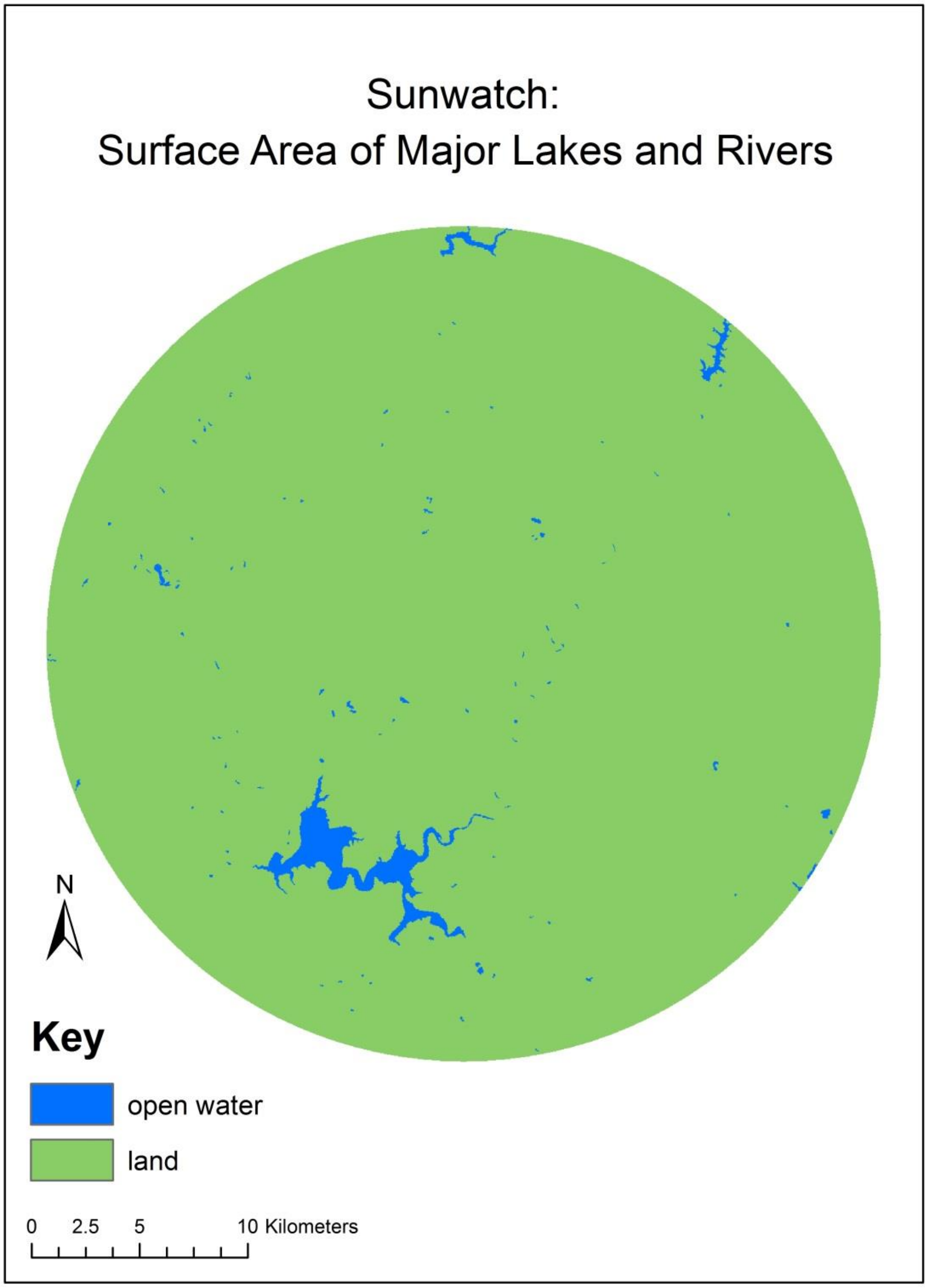

Figure 127. Lakes and rivers at Sunwatch.

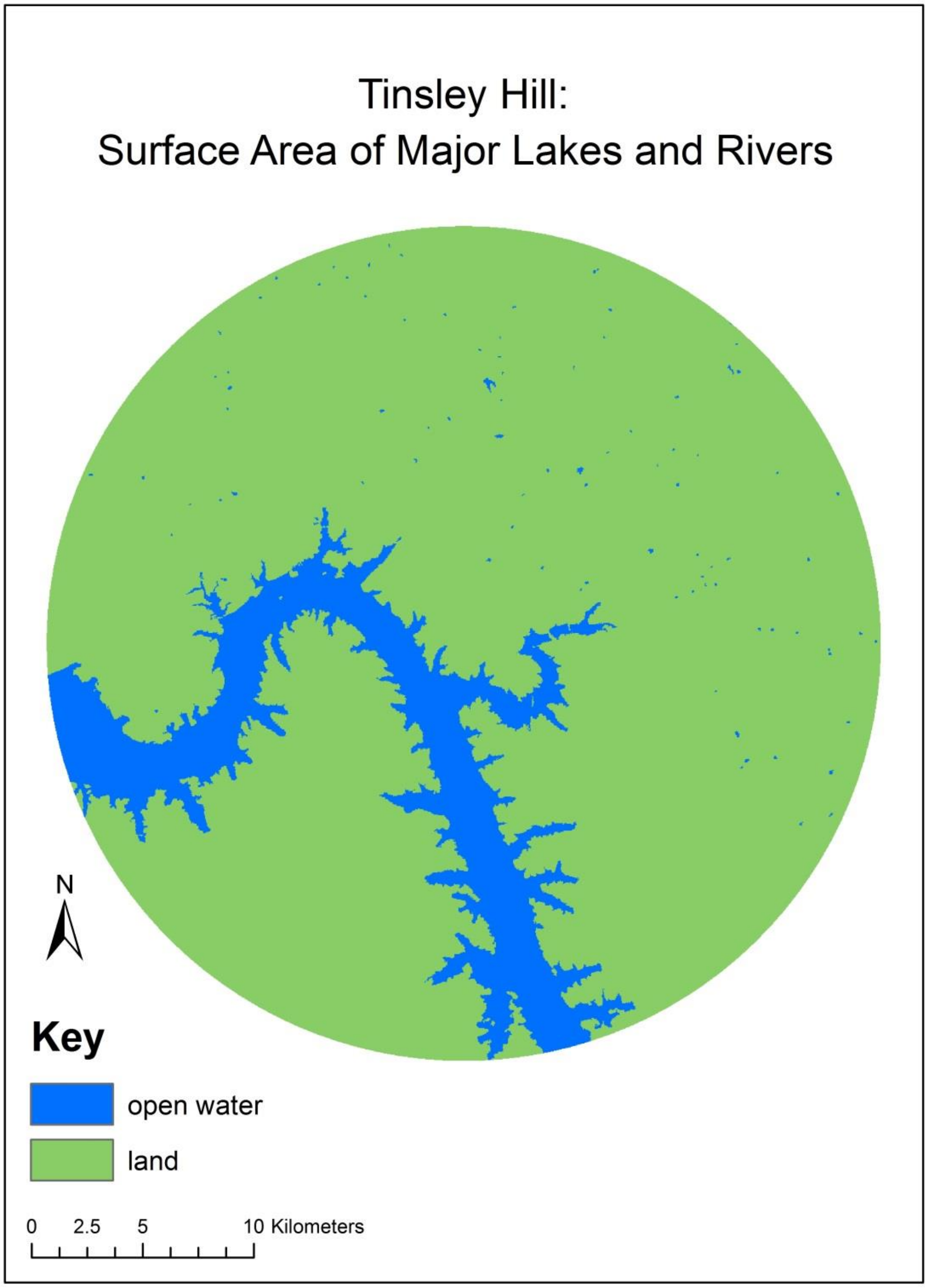

Figure 128. Lakes and rivers at Tinsley Hill.

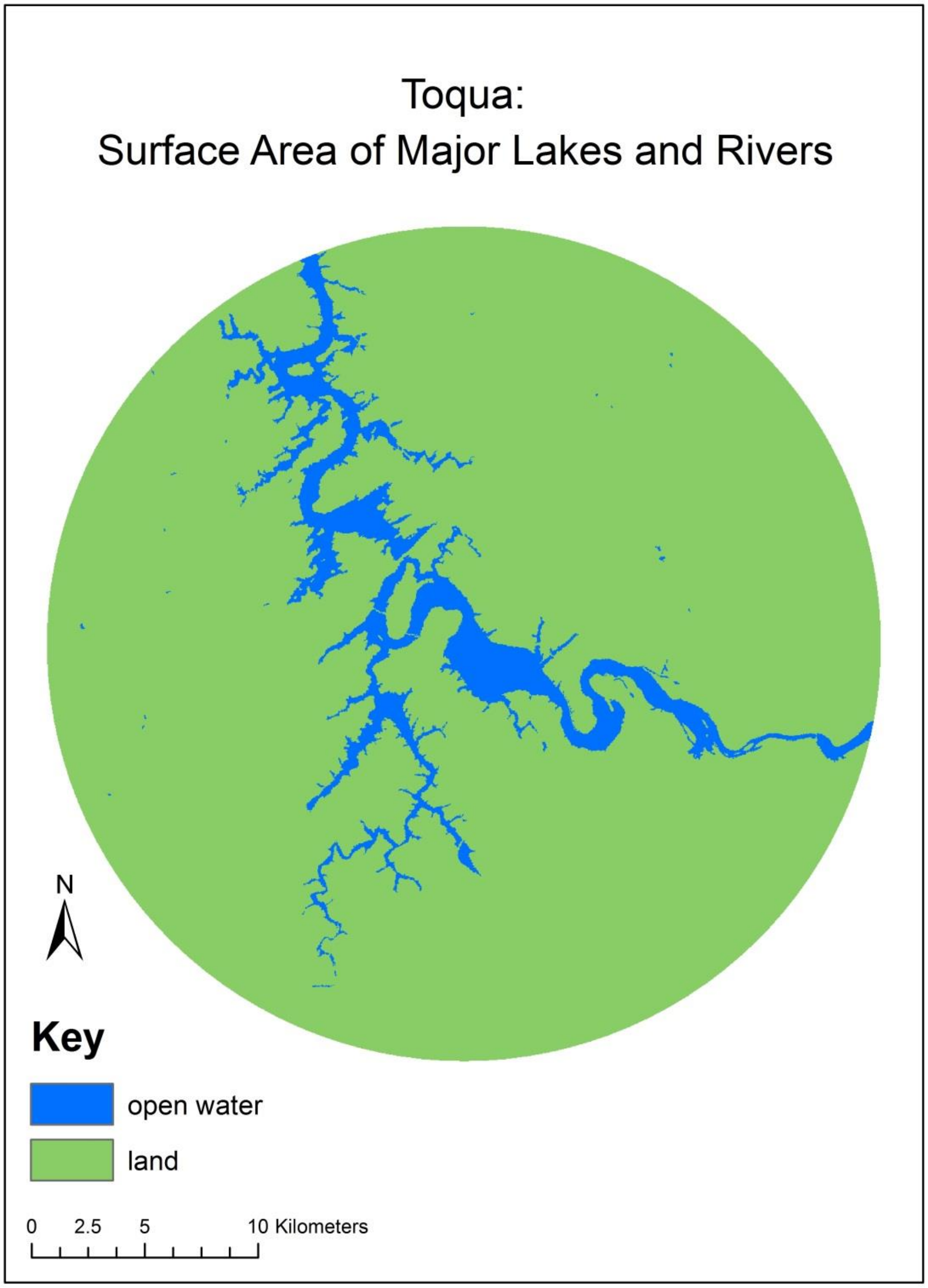

Figure 129. Lakes and rivers at Toqua.

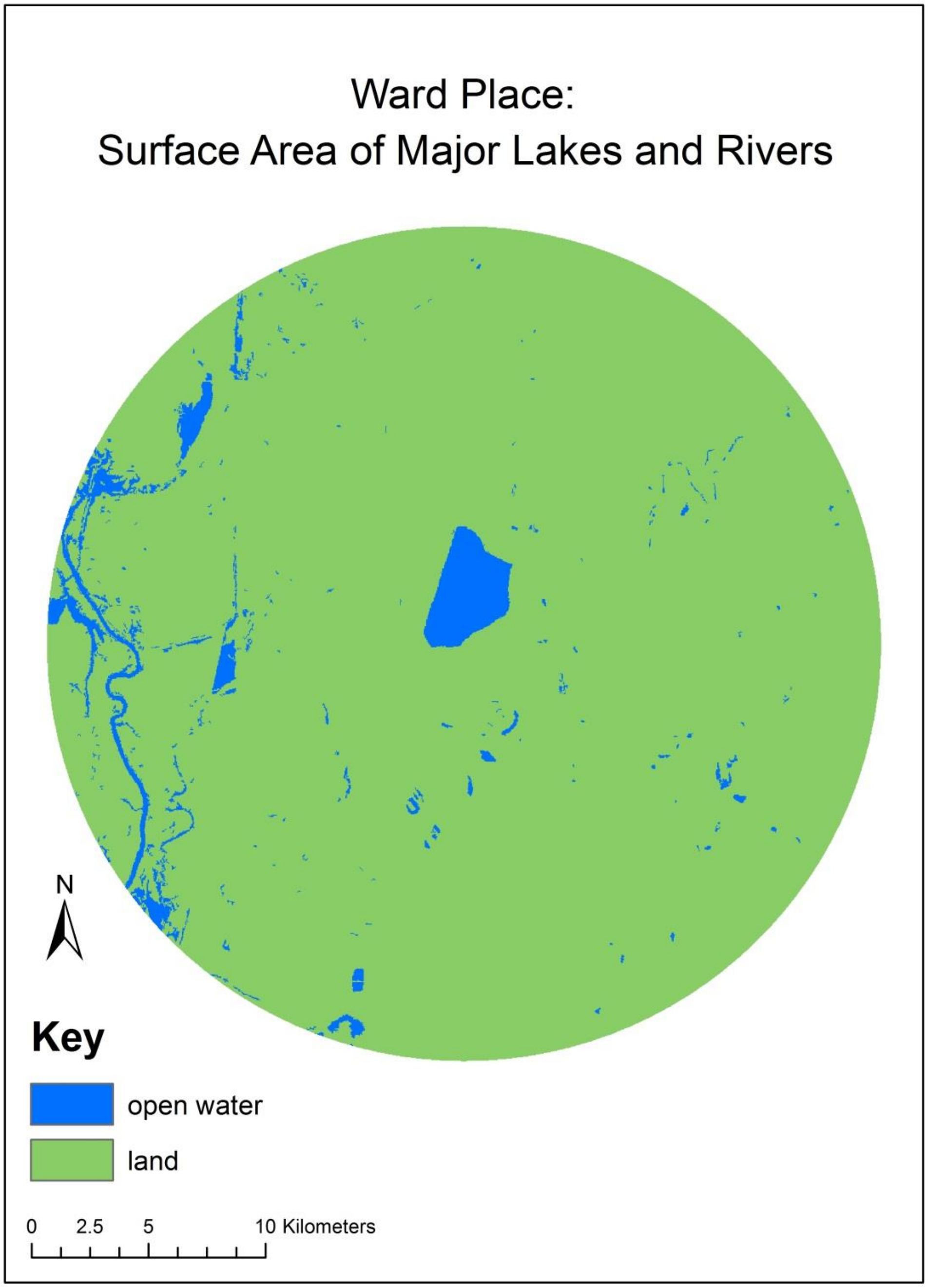

Figure 130. Lakes and rivers at Ward Place.

APPENDIX B

Raw data used for analysis

Table 15. Human data used for statistical analysis.

Site name	Overall rate % (*n*)	Adult rate % (*n*)	Child rate % (*n*)	Male rate % (*n*)	Female rate % (*n*)	PH rate % (*n*)	CO rate % (*n*)	PH rate, adults % (*n*)	CO rate, adults % (*n*)	PH rate, children % (*n*)	CO rate, children % (*n*)	Historic rate % (*n*)	$\delta^{13}C$ average (*n*)
Irene Mound	2.67 (187)	2.65 (151)	5.56 (36)	1.82 (55)	3.75 (80)	0.54 (186)	2.86 (175)	0.67 (150)	2.14 (140)	0.00 (36)	5.71 (35)		-13.31 (10)
Monongahela	28.10 (121)	9.09 (66)	47.27 (55)	8.82 (34)	9.38 (32)	17.50 (120)	16.00 (100)	6.15 (65)	7.02 (57)	30.91 (55)	27.91 (43)		-10.50 (11)
Buffalo	13.13 (99)	12.96 (54)	11.11 (45)	12.00 (25)	13.79 (29)	12.12 (99)	2.20 (91)	14.81 (54)	1.92 (52)	8.89 (45)	2.56 (39)		
Pearson	3.16 (95)	6.52 (46)	0.00 (50)	8.70 (23)	4.35 (23)	3.16 (95)	0.00 (69)	6.52 (46)	0.00 (29)	0.00 (49)	0.00 (29)		-12.81 (4)
Sunwatch	19.53 (128)	16.36 (55)	21.62 (74)	12.90 (31)	20.83 (24)	15.87 (126)	8.62 (116)	16.67 (72)	1.89 (63)	15.28 (72)	14.29 (63)		-11.10 (58)
Boytt's Field	4.00 (25)	4.00 (25)										44.51 (1,846)	
Ward Place	0.00 (25)	0.00 (25)										18.37 (283)	
Mount Nebo	0.00 (77)	0.00 (54)	0.00 (23)									1.62 (1,910)	
Anderson	65.91 (44)	75.86 (29)	87.50 (8)	52.63 (19)	70.59 (17)								-10.60 (8)
Ledford Island	20.41 (343)	22.90 (214)	16.28 (129)									28.40 (764)	
Moundville	9.26 (162)	4.35 (115)	21.28 (47)			0.00 (162)	9.26 (162)						-10.80 (37)
East St. Louis Stone Quarry	17.07 (41)	12.90 (31)	30.00 (10)										-10.96 (21)
Kane Mounds	6.12 (98)	4.05 (74)	12.50 (24)										-10.30 (4)
Averbuch	39.07 (732)	42.92 (459)	32.60 (273)	46.85 (222)	43.78 (201)								-8.00 (4)
Norris Farms #36	44.71 (170)	15.79 (95)	81.33 (75)										-12.60 (5)
Hardin Village	8.22 (292)	7.14 (140)	9.21 (152)	4.48 (67)	9.59 (73)							19.23 (208)	-11.64 (49)
Eiden	17.21 (122)	5.49 (91)	51.61 (31)	2.33 (43)	8.33 (48)	9.84 (122)	17.07 (123)						

Table 15 continued.

Tinsley Hill	2.47 (81)											1.44 (277)	-8.60 (19)
Lewis Creek Mound	11.54 (26)	15.38 (13)	7.69 (13)			15.38 (13)	7.69 (13)	15.38 (13)	0.00 (0)	0.00 (0)	7.69 (13)	18 (451)	-12.38 (9)
Cox	27.37 (190)	22.14 (131)	38.98 (59)	18.84 (69)	25.81 (62)	13.44 (186)	21.74 (138)	13.85 (130)	13.21 (106)	12.50 (56)	51.56 (33)	22.17 (212)	
Etowah	3.20 (125)	3.20 (125)										6.90 (?)	
Toqua	24.49 (245)	17.91 (134)	32.43 (111)	17.07 (82)	23.19 (69)							29.98 (4,817)	
Juhle	17.79 (208)	6.94 (144)	42.19 (64)	4.92 (61)	8.54 (82)	10.58 (208)	15.27 (203)	3.50 (143)	4.86 (144)	26.15 (65)	37.29 (59)		

Table 16. Environmental data used for statistical analysis.

Site name	longitude	latitude	Well-drained soil (%)	Elevation (m above sea level)	Average annual minimum temperature (°F)	Average annual precipitation (mm)	Land covered by major lakes and rivers (%)
Irene Mound	-81.150000	32.140000	10.31	16	55.7	48.6	4.61
Monongahela	-79.270000	40.090000	91.03	628	37.0	48.4	0.28
Buffalo	-81.300000	38.210000	99.98	403	42.1	44.8	1.32
Pearson	-83.041200	41.134800	17.58	253	39.9	37.0	0.53
SunWatch	-84.074800	39.090000	45.12	267	43.4	42.0	1.56
Boytt's Field	-92.373075	33.280171	37.27	38	50.4	54.4	1.60
Ward Place	-91.930604	32.845774	31.64	34	52.2	56.5	3.66
Mount Nebo	-91.438000	32.297110	8.25	25	53.1	56.1	0.53
Anderson	-84.097560	39.449880	75.45	268	42.1	40.6	2.08
Ledford Island	-84.836700	35.333820	96.58	252	46.4	53.8	3.28
Moundville	-87.627265	33.002995	74.85	77	51.0	53.8	2.66
East St. Louis Stone Quarry	-90.182160	38.540100	51.78	150	46.0	42.2	3.76
Kane Mounds	-90.014275	38.759150	52.41	143	45.9	41.0	5.49
Averbuch	-86.848755	36.223933	98.10	185	45.8	49.6	1.84
Norris Farms #36	-90.089260	40.374170	64.56	158	41.0	38.1	7.41
Hardin Village	-82.903525	38.749765	97.08	219	41.3	40.2	2.64
Eiden	-82.112150	41.459250	9.91	190	42.4	36.9	40.27
Tinsley Hill	-88.042430	37.033805	97.68	142	47.4	48.6	12.18
Lewis Creek Mound	-78.988730	38.174815	88.77	420	41.5	38.3	0.33
Cox	-84.159420	36.010680	98.02	308	46.8	50.9	3.03
Etowah	-84.806550	34.127816	96.56	262	48.7	50.0	4.12
Toqua	-84.189805	35.575940	99.27	303	46.3	51.6	7.03
Juhle	-77.152338	38.455366	76.11	22	46.6	41.3	31.19

APPENDIX C

Linear regression output from SPSS

C1. Multiple linear regression, adults, five environmental variables

Descriptive Statistics

	Mean	Std. Deviation	N
SQRAdult	.2780	.13625	20
TempMin	45.885	4.8993	20
Precip	46.295	6.8240	20
SoilDrain	.6235390	.33981595	20
Elev	208.400	159.0380	20
Water	.0626460	.10390770	20

Correlations

		SQRAdult	TempMin	Precip	SoilDrain	Elev	Water
Pearson Correlation	SQRAdult	1.000	-.562	-.332	.612	.569	-.047
	TempMin	-.562	1.000	.690	-.368	-.752	-.072
	Precip	-.332	.690	1.000	.055	-.225	-.367
	SoilDrain	.612	-.368	.055	1.000	.586	-.212
	Elev	.569	-.752	-.225	.586	1.000	-.250
	Water	-.047	-.072	-.367	-.212	-.250	1.000
Sig. (1-tailed)	SQRAdult	.	.005	.076	.002	.004	.421
	TempMin	.005	.	.000	.055	.000	.381
	Precip	.076	.000	.	.408	.171	.056
	SoilDrain	.002	.055	.408	.	.003	.185
	Elev	.004	.000	.171	.003	.	.144
	Water	.421	.381	.056	.185	.144	.
N	SQRAdult	20	20	20	20	20	20
	TempMin	20	20	20	20	20	20
	Precip	20	20	20	20	20	20
	SoilDrain	20	20	20	20	20	20
	Elev	20	20	20	20	20	20
	Water	20	20	20	20	20	20

Variables Entered/Removed[a]

Model	Variables Entered	Variables Removed	Method
1	Water, TempMin, SoilDrain, Precip, Elev[b]	.	Enter

a. Dependent Variable: SQRAdult

b. All requested variables entered.

Model Summary[b]

Model	R	R Square	Adjusted R Square	Std. Error of the Estimate
1	.730[a]	.532	.365	.10855

a. Predictors: (Constant), Water, TempMin, SoilDrain, Precip, Elev

b. Dependent Variable: SQRAdult

ANOVA[a]

Model		Sum of Squares	df	Mean Square	F	Sig.
1	Regression	.188	5	.038	3.187	.040[b]
	Residual	.165	14	.012		
	Total	.353	19			

a. Dependent Variable: SQRAdult

b. Predictors: (Constant), Water, TempMin, SoilDrain, Precip, Elev

Coefficients[a]

Model		Unstandardized Coefficients B	Unstandardized Coefficients Std. Error	Standardized Coefficients Beta	t	Sig.	95.0% Confidence Interval for B Lower Bound	95.0% Confidence Interval for B Upper Bound	Correlations Zero-order	Correlations Partial	Correlations Part	Collinearity Statistics Tolerance	Collinearity Statistics VIF
1	(Constant)	.448	.483		.927	.370	-.589	1.484					
	TempMin	-.001	.013	-.042	-.087	.932	-.030	.027	-.562	-.023	-.016	.146	6.863
	Precip	-.006	.007	-.297	-.879	.394	-.020	.009	-.332	-.229	-.161	.294	3.404
	SoilDrain	.205	.094	.512	2.196	.045	.005	.406	.612	.506	.401	.614	1.629
	Elev	.000	.000	.169	.439	.668	-.001	.001	.569	.116	.080	.225	4.443
	Water	-.011	.278	-.008	-.040	.969	-.607	.585	-.047	-.011	-.007	.744	1.344

a. Dependent Variable: SQRAdult

Coefficient Correlations[a]

Model			Water	TempMin	SoilDrain	Precip	Elev
1	Correlations	Water	1.000	.082	-.018	.218	.263
		TempMin	.082	1.000	.096	-.770	.761
		SoilDrain	-.018	.096	1.000	-.216	-.310
		Precip	.218	-.770	-.216	1.000	-.416
		Elev	.263	.761	-.310	-.416	1.000
	Covariances	Water	.077	.000	.000	.000	2.409E-5
		TempMin	.000	.000	.000	-6.902E-5	3.342E-6
		SoilDrain	.000	.000	.009	.000	-9.556E-6
		Precip	.000	-6.902E-5	.000	4.533E-5	-9.253E-7
		Elev	2.409E-5	3.342E-6	-9.556E-6	-9.253E-7	1.089E-7

a. Dependent Variable: SQRAdult

Collinearity Diagnostics[a]

Model	Dimension	Eigenvalue	Condition Index	Variance Proportions (Constant)	TempMin	Precip	SoilDrain	Elev	Water
1	1	4.786	1.000	.00	.00	.00	.01	.00	.01
	2	.795	2.453	.00	.00	.00	.01	.01	.56
	3	.307	3.946	.00	.00	.00	.03	.12	.17
	4	.103	6.820	.00	.00	.00	.92	.20	.00
	5	.007	26.121	.14	.01	.49	.03	.05	.24
	6	.001	71.565	.86	.99	.51	.01	.62	.02

a. Dependent Variable: SQRAdult

Residuals Statistics[a]

	Minimum	Maximum	Mean	Std. Deviation	N
Predicted Value	.0744	.4160	.2780	.09940	20
Std. Predicted Value	-2.047	1.388	.000	1.000	20
Standard Error of Predicted Value	.037	.096	.057	.017	20
Adjusted Predicted Value	.0614	.5317	.2845	.11893	20
Residual	-.15213	.16863	.00000	.09318	20
Std. Residual	-1.401	1.553	.000	.858	20
Stud. Residual	-1.585	1.733	-.026	.996	20
Deleted Residual	-.23020	.20981	-.00657	.12890	20
Stud. Deleted Residual	-1.687	1.884	-.027	1.029	20
Mahal. Distance	1.240	13.806	4.750	3.447	20
Cook's Distance	.000	.441	.067	.098	20
Centered Leverage Value	.065	.727	.250	.181	20

a. Dependent Variable: SQRAdult

C2. Multiple linear regression, children, five environmental variables

Descriptive Statistics

	Mean	Std. Deviation	N
SQRKid	.4236	.21692	17
SoilDrain	.6559759	.35612013	17
Elev	227.118	160.1358	17
TempMin	45.365	4.7396	17
Precip	45.676	6.3170	17
Water	.0649094	.11279424	17

Correlations

		SQRKid	SoilDrain	Elev	TempMin	Precip	Water
Pearson Correlation	SQRKid	1.000	.418	.239	-.226	.002	.492
	SoilDrain	.418	1.000	.514	-.339	.223	-.267
	Elev	.239	.514	1.000	-.782	-.121	-.307
	TempMin	-.226	-.339	-.782	1.000	.587	-.021
	Precip	.002	.223	-.121	.587	1.000	-.372
	Water	.492	-.267	-.307	-.021	-.372	1.000
Sig. (1-tailed)	SQRKid	.	.047	.178	.192	.497	.022
	SoilDrain	.047	.	.017	.091	.194	.150
	Elev	.178	.017	.	.000	.322	.115
	TempMin	.192	.091	.000	.	.007	.467
	Precip	.497	.194	.322	.007	.	.071
	Water	.022	.150	.115	.467	.071	.
N	SQRKid	17	17	17	17	17	17
	SoilDrain	17	17	17	17	17	17
	Elev	17	17	17	17	17	17
	TempMin	17	17	17	17	17	17
	Precip	17	17	17	17	17	17
	Water	17	17	17	17	17	17

Variables Entered/Removed[a]

Model	Variables Entered	Variables Removed	Method
1	Water, TempMin, SoilDrain, Precip, Elev[b]	.	Enter

a. Dependent Variable: SQRKid

b. All requested variables entered.

Model Summary[b]

Model	R	R Square	Adjusted R Square	Std. Error of the Estimate
1	.798[a]	.637	.472	.15761

a. Predictors: (Constant), Water, TempMin, SoilDrain, Precip, Elev

b. Dependent Variable: SQRKid

ANOVA[a]

Model		Sum of Squares	df	Mean Square	F	Sig.
1	Regression	.480	5	.096	3.862	.029[b]
	Residual	.273	11	.025		
	Total	.753	16			

a. Dependent Variable: SQRKid

b. Predictors: (Constant), Water, TempMin, SoilDrain, Precip, Elev

Coefficients[a]

Model		Unstandardized Coefficients		Standardized Coefficients	t	Sig.	95.0% Confidence Interval for B		Correlations			Collinearity Statistics	
		B	Std. Error	Beta			Lower Bound	Upper Bound	Zero-order	Partial	Part	Tolerance	VIF
1	(Constant)	-.585	.865		-.676	.513	-2.488	1.319					
	SoilDrain	.274	.142	.450	1.931	.080	-.038	.587	.418	.503	.351	.607	1.647
	Elev	.001	.001	.413	.990	.343	-.001	.002	.239	.286	.180	.190	5.266
	TempMin	.008	.023	.185	.365	.722	-.043	.059	-.226	.110	.066	.129	7.757
	Precip	.005	.011	.139	.419	.683	-.020	.030	.002	.125	.076	.299	3.347
	Water	1.530	.414	.795	3.694	.004	.618	2.441	.492	.744	.671	.712	1.405

a. Dependent Variable: SQRKid

Coefficient Correlations[a]

Model			Water	TempMin	SoilDrain	Precip	Elev
1	Correlations	Water	1.000	.179	.034	.126	.331
		TempMin	.179	1.000	.263	-.766	.828
		SoilDrain	.034	.263	1.000	-.394	-.071
		Precip	.126	-.766	-.394	1.000	-.497
		Elev	.331	.828	-.071	-.497	1.000
	Covariances	Water	.171	.002	.002	.001	7.731E-5
		TempMin	.002	.001	.001	.000	1.082E-5
		SoilDrain	.002	.001	.020	-.001	-5.668E-6
		Precip	.001	.000	-.001	.000	-3.200E-6
		Elev	7.731E-5	1.082E-5	-5.668E-6	-3.200E-6	3.188E-7

a. Dependent Variable: SQRKid

Collinearity Diagnostics[a]

			Condition	Variance Proportions					
Model	Dimension	Eigenvalue	Index	(Constant)	SoilDrain	Elev	TempMin	Precip	Water
1	1	4.799	1.000	.00	.00	.00	.00	.00	.01
	2	.824	2.413	.00	.01	.01	.00	.00	.54
	3	.255	4.337	.00	.03	.12	.00	.00	.19
	4	.115	6.472	.00	.82	.12	.00	.00	.01
	5	.006	27.372	.11	.07	.03	.01	.49	.20
	6	.001	79.583	.89	.07	.72	.99	.50	.05

a. Dependent Variable: SQRKid

Residuals Statistics[a]

	Minimum	Maximum	Mean	Std. Deviation	N
Predicted Value	.1276	.7051	.4236	.17314	17
Std. Predicted Value	-1.710	1.626	.000	1.000	17
Standard Error of Predicted Value	.057	.138	.090	.027	17
Adjusted Predicted Value	.2110	.7653	.4379	.15941	17
Residual	-.17741	.25813	.00000	.13068	17
Std. Residual	-1.126	1.638	.000	.829	17
Stud. Residual	-1.651	1.758	-.040	1.016	17
Deleted Residual	-.38176	.30240	-.01434	.20484	17
Stud. Deleted Residual	-1.815	1.977	-.039	1.067	17
Mahal. Distance	1.175	11.255	4.706	3.274	17
Cook's Distance	.002	.523	.104	.150	17
Centered Leverage Value	.073	.703	.294	.205	17

a. Dependent Variable: SQRKid

C3. Multiple linear regression, PH in children, four environmental variables

Descriptive Statistics

	Mean	Std. Deviation	N
AllPHProp	.117163	.1201099	8
SoilDrain	.6586525	.36466248	8
TempMin	44.125	5.6996	8
Precip	43.913	5.0919	8
Water	.0535600	.10545824	8

Correlations

		AllPHProp	SoilDrain	TempMin	Precip	Water
Pearson Correlation	AllPHProp	1.000	.468	-.315	.319	.446
	SoilDrain	.468	1.000	-.427	.251	.060
	TempMin	-.315	-.427	1.000	.384	.308
	Precip	.319	.251	.384	1.000	-.119
	Water	.446	.060	.308	-.119	1.000
Sig. (1-tailed)	AllPHProp	.	.121	.223	.221	.134
	SoilDrain	.121	.	.146	.275	.443
	TempMin	.223	.146	.	.174	.229
	Precip	.221	.275	.174	.	.389
	Water	.134	.443	.229	.389	.
N	AllPHProp	8	8	8	8	8
	SoilDrain	8	8	8	8	8
	TempMin	8	8	8	8	8
	Precip	8	8	8	8	8
	Water	8	8	8	8	8

Variables Entered/Removed[a]

Model	Variables Entered	Variables Removed	Method
1	Water, SoilDrain, Precip, TempMin[b]	.	Enter

a. Dependent Variable: AllPHProp

b. All requested variables entered.

Model Summary[b]

Model	R	R Square	Adjusted R Square	Std. Error of the Estimate
1	.942[a]	.887	.735	.0618098

a. Predictors: (Constant), Water, SoilDrain, Precip, TempMin

b. Dependent Variable: AllPHProp

ANOVA[a]

Model		Sum of Squares	df	Mean Square	F	Sig.
1	Regression	.090	4	.022	5.858	.089[b]
	Residual	.011	3	.004		
	Total	.101	7			

a. Dependent Variable: AllPHProp

b. Predictors: (Constant), Water, SoilDrain, Precip, TempMin

Coefficients[a]

Model		Unstandardized Coefficients		Standardized Coefficients	t	Sig.	95.0% Confidence Interval for B		Correlations			Collinearity Statistics	
		B	Std. Error	Beta			Lower Bound	Upper Bound	Zero-order	Partial	Part	Tolerance	VIF
1	(Constant)	.163	.246		.665	.554	-.618	.945					
	SoilDrain	-.082	.090	-.248	-.906	.432	-.369	.206	.468	-.464	-.176	.503	1.987
	TempMin	-.022	.007	-1.034	-3.332	.045	-.043	-.001	-.315	-.887	-.648	.393	2.545
	Precip	.021	.006	.883	3.250	.047	.000	.041	.319	.882	.632	.512	1.954
	Water	1.008	.268	.885	3.768	.033	.157	1.860	.446	.909	.733	.685	1.460

a. Dependent Variable: AllPHProp

Coefficient Correlations[a]

Model			Water	SoilDrain	Precip	TempMin
1	Correlations	Water	1.000	-.428	.451	-.547
		SoilDrain	-.428	1.000	-.593	.677
		Precip	.451	-.593	1.000	-.666
		TempMin	-.547	.677	-.666	1.000
	Covariances	Water	.072	-.010	.001	-.001
		SoilDrain	-.010	.008	.000	.000
		Precip	.001	.000	4.113E-5	-2.791E-5
		TempMin	-.001	.000	-2.791E-5	4.275E-5

a. Dependent Variable: AllPHProp

Collinearity Diagnostics[a]

Model	Dimension	Eigenvalue	Condition Index	Variance Proportions				
				(Constant)	SoilDrain	TempMin	Precip	Water
1	1	4.102	1.000	.00	.01	.00	.00	.01
	2	.712	2.400	.00	.00	.00	.00	.68
	3	.177	4.820	.00	.45	.00	.00	.00
	4	.006	26.745	.87	.00	.05	.34	.00
	5	.003	35.128	.13	.54	.95	.66	.31

a. Dependent Variable: AllPHProp

Residuals Statistics[a]

	Minimum	Maximum	Mean	Std. Deviation	N
Predicted Value	-.012045	.294141	.117162	.1130887	8
Std. Predicted Value	-1.143	1.565	.000	1.000	8
Standard Error of Predicted Value	.028	.062	.048	.012	8
Adjusted Predicted Value	-1.758297	.241687	-.127575	.6658571	8
Residual	-.0560534	.0810503	.0000000	.0404640	8
Std. Residual	-.907	1.311	.000	.655	8
Stud. Residual	-1.621	1.468	.019	.933	8
Deleted Residual	-.1790083	2.0197971	.2447376	.7224096	8
Stud. Deleted Residual	-3.750	2.258	-.156	1.688	8
Mahal. Distance	.539	6.123	3.500	1.932	8
Cook's Distance	.001	213.504	26.888	75.405	8
Centered Leverage Value	.077	.875	.500	.276	8

a. Dependent Variable: AllPHProp

C4. Multiple linear regression, CO in children, four environmental variables

Descriptive Statistics

	Mean	Std. Deviation	N
AllCOProp	.183765	.1862756	8
SoilDrain	.6586525	.36466248	8
TempMin	44.125	5.6996	8
Precip	43.913	5.0919	8
Water	.0535600	.10545824	8

Correlations

		AllCOProp	SoilDrain	TempMin	Precip	Water
Pearson Correlation	AllCOProp	1.000	.503	.083	.522	.432
	SoilDrain	.503	1.000	-.427	.251	.060
	TempMin	.083	-.427	1.000	.384	.308
	Precip	.522	.251	.384	1.000	-.119
	Water	.432	.060	.308	-.119	1.000
Sig. (1-tailed)	AllCOProp	.	.102	.423	.092	.142
	SoilDrain	.102	.	.146	.275	.443
	TempMin	.423	.146	.	.174	.229
	Precip	.092	.275	.174	.	.389
	Water	.142	.443	.229	.389	.
N	AllCOProp	8	8	8	8	8
	SoilDrain	8	8	8	8	8
	TempMin	8	8	8	8	8
	Precip	8	8	8	8	8
	Water	8	8	8	8	8

Variables Entered/Removed[a]

Model	Variables Entered	Variables Removed	Method
1	Water, SoilDrain, Precip, TempMin[b]	.	Enter

a. Dependent Variable: AllCOProp

b. All requested variables entered.

Model Summary[b]

Model	R	R Square	Adjusted R Square	Std. Error of the Estimate
1	.813[a]	.661	.209	.1656863

a. Predictors: (Constant), Water, SoilDrain, Precip, TempMin

b. Dependent Variable: AllCOProp

ANOVA[a]

Model		Sum of Squares	df	Mean Square	F	Sig.
1	Regression	.161	4	.040	1.462	.393[b]
	Residual	.082	3	.027		
	Total	.243	7			

a. Dependent Variable: AllCOProp

b. Predictors: (Constant), Water, SoilDrain, Precip, TempMin

Coefficients[a]

Model		Unstandardized Coefficients		Standardized Coefficients			95.0% Confidence Interval for B		Correlations			Collinearity Statistics	
		B	Std. Error	Beta	t	Sig.	Lower Bound	Upper Bound	Zero-order	Partial	Part	Tolerance	VIF
1	(Constant)	-.597	.658		-.907	.431	-2.692	1.498					
	SoilDrain	.102	.242	.200	.423	.701	-.668	.873	.503	.237	.142	.503	1.987
	TempMin	-.008	.018	-.253	-.472	.669	-.064	.047	.083	-.263	-.159	.393	2.545
	Precip	.023	.017	.638	1.357	.268	-.031	.078	.522	.617	.456	.512	1.954
	Water	1.014	.717	.574	1.414	.252	-1.269	3.297	.432	.632	.475	.685	1.460

a. Dependent Variable: AllCOProp

Coefficient Correlations[a]

Model			Water	SoilDrain	Precip	TempMin
1	Correlations	Water	1.000	-.428	.451	-.547
		SoilDrain	-.428	1.000	-.593	.677
		Precip	.451	-.593	1.000	-.666
		TempMin	-.547	.677	-.666	1.000
	Covariances	Water	.515	-.074	.006	-.007
		SoilDrain	-.074	.059	-.002	.003
		Precip	.006	-.002	.000	.000
		TempMin	-.007	.003	.000	.000

a. Dependent Variable: AllCOProp

Collinearity Diagnostics[a]

Model	Dimension	Eigenvalue	Condition Index	Variance Proportions				
				(Constant)	SoilDrain	TempMin	Precip	Water
1	1	4.102	1.000	.00	.01	.00	.00	.01
	2	.712	2.400	.00	.00	.00	.00	.68
	3	.177	4.820	.00	.45	.00	.00	.00
	4	.006	26.745	.87	.00	.05	.34	.00
	5	.003	35.128	.13	.54	.95	.66	.31

a. Dependent Variable: AllCOProp

Residuals Statistics[a]

	Minimum	Maximum	Mean	Std. Deviation	N
Predicted Value	-.040843	.374885	.183765	.1514380	8
Std. Predicted Value	-1.483	1.262	.000	1.000	8
Standard Error of Predicted Value	.074	.166	.128	.032	8
Adjusted Predicted Value	-.130435	7.316087	1.095534	2.5251097	8
Residual	-.1896029	.1816277	.0000000	.1084671	8
Std. Residual	-1.144	1.096	.000	.655	8
Stud. Residual	-1.372	1.506	-.147	.978	8
Deleted Residual	-6.9431868	.3426032	-.9117685	2.4524152	8
Stud. Deleted Residual	-1.837	2.486	-.108	1.326	8
Mahal. Distance	.539	6.123	3.500	1.932	8
Cook's Distance	.008	351.116	44.225	124.004	8

Centered Leverage Value	.077	.875	.500	.276	8

a. Dependent Variable: AllCOProp

C5. Multiple linear regression, PH in adults, four environmental variables

Descriptive Statistics

	Mean	Std. Deviation	N
AllPHProp	.096943	.0617513	8
SoilDrain	.6586525	.36466248	8
TempMin	44.125	5.6996	8
Precip	43.913	5.0919	8
Water	.0535600	.10545824	8

Correlations

		AllPHProp	SoilDrain	TempMin	Precip	Water
Pearson Correlation	AllPHProp	1.000	.505	-.407	-.144	-.451
	SoilDrain	.505	1.000	-.427	.251	.060
	TempMin	-.407	-.427	1.000	.384	.308
	Precip	-.144	.251	.384	1.000	-.119
	Water	-.451	.060	.308	-.119	1.000
Sig. (1-tailed)	AllPHProp	.	.101	.158	.366	.131
	SoilDrain	.101	.	.146	.275	.443
	TempMin	.158	.146	.	.174	.229
	Precip	.366	.275	.174	.	.389
	Water	.131	.443	.229	.389	.
N	AllPHProp	8	8	8	8	8
	SoilDrain	8	8	8	8	8
	TempMin	8	8	8	8	8
	Precip	8	8	8	8	8
	Water	8	8	8	8	8

Variables Entered/Removed[a]

Model	Variables Entered	Variables Removed	Method
1	Water, SoilDrain, Precip, TempMin[b]	.	Enter

a. Dependent Variable: AllPHProp

b. All requested variables entered.

Model Summary[b]

Model	R	R Square	Adjusted R Square	Std. Error of the Estimate
1	.824[a]	.679	.252	.0534183

a. Predictors: (Constant), Water, SoilDrain, Precip, TempMin

b. Dependent Variable: AllPHProp

ANOVA[a]

Model		Sum of Squares	df	Mean Square	F	Sig.
1	Regression	.018	4	.005	1.589	.367[b]
	Residual	.009	3	.003		
	Total	.027	7			

a. Dependent Variable: AllPHProp

b. Predictors: (Constant), Water, SoilDrain, Precip, TempMin

Coefficients[a]

Model		Unstandardized Coefficients		Standardized Coefficients	t	Sig.	95.0% Confidence Interval for B		Correlations			Collinearity Statistics	
		B	Std. Error	Beta			Lower Bound	Upper Bound	Zero-order	Partial	Part	Tolerance	VIF
1	(Constant)	.145	.212		.683	.544	-.531	.821					
	SoilDrain	.149	.078	.880	1.910	.152	-.099	.397	.505	.741	.624	.503	1.987
	TempMin	.005	.006	.421	.806	.479	-.013	.023	-.407	.422	.264	.393	2.545
	Precip	-.007	.006	-.611	-1.336	.274	-.025	.010	-.144	-.611	-.437	.512	1.954
	Water	-.414	.231	-.706	-1.788	.172	-1.150	.323	-.451	-.718	-.585	.685	1.460

a. Dependent Variable: AllPHProp

Coefficient Correlations[a]

Model			Water	SoilDrain	Precip	TempMin
1	Correlations	Water	1.000	-.428	.451	-.547
		SoilDrain	-.428	1.000	-.593	.677
		Precip	.451	-.593	1.000	-.666
		TempMin	-.547	.677	-.666	1.000
	Covariances	Water	.053	-.008	.001	-.001
		SoilDrain	-.008	.006	.000	.000
		Precip	.001	.000	3.072E-5	-2.085E-5
		TempMin	-.001	.000	-2.085E-5	3.193E-5

a. Dependent Variable: AllPHProp

Collinearity Diagnostics[a]

			Condition	Variance Proportions				
Model	Dimension	Eigenvalue	Index	(Constant)	SoilDrain	TempMin	Precip	Water
1	1	4.102	1.000	.00	.01	.00	.00	.01
	2	.712	2.400	.00	.00	.00	.00	.68
	3	.177	4.820	.00	.45	.00	.00	.00
	4	.006	26.745	.87	.00	.05	.34	.00
	5	.003	35.128	.13	.54	.95	.66	.31

a. Dependent Variable: AllPHProp

Residuals Statistics[a]

	Minimum	Maximum	Mean	Std. Deviation	N
Predicted Value	.035297	.181457	.096943	.0508949	8
Std. Predicted Value	-1.211	1.661	.000	1.000	8
Standard Error of Predicted Value	.024	.053	.041	.010	8
Adjusted Predicted Value	.073859	3.517408	.572388	1.1914826	8
Residual	-.0285974	.0740906	.0000000	.0349705	8
Std. Residual	-.535	1.387	.000	.655	8
Stud. Residual	-1.419	1.553	-.354	1.022	8
Deleted Residual	-3.4824083	.0928406	-.4754444	1.2186536	8
Stud. Deleted Residual	-2.018	2.860	-.282	1.496	8

Mahal. Distance	.539	6.123	3.500	1.932	8
Cook's Distance	.000	849.738	106.715	300.228	8
Centered Leverage Value	.077	.875	.500	.276	8

a. Dependent Variable: AllPHProp

C6. Multiple linear regression, CO in adults, four environmental variables

Descriptive Statistics

	Mean	Std. Deviation	N
AllCOProp	.038800	.0445304	8
SoilDrain	.6586525	.36466248	8
TempMin	44.125	5.6996	8
Precip	43.913	5.0919	8
Water	.0535600	.10545824	8

Correlations

		AllCOProp	SoilDrain	TempMin	Precip	Water
Pearson Correlation	AllCOProp	1.000	.491	.095	.751	.130
	SoilDrain	.491	1.000	-.427	.251	.060
	TempMin	.095	-.427	1.000	.384	.308
	Precip	.751	.251	.384	1.000	-.119
	Water	.130	.060	.308	-.119	1.000
Sig. (1-tailed)	AllCOProp	.	.108	.412	.016	.380
	SoilDrain	.108	.	.146	.275	.443
	TempMin	.412	.146	.	.174	.229
	Precip	.016	.275	.174	.	.389
	Water	.380	.443	.229	.389	.
N	AllCOProp	8	8	8	8	8
	SoilDrain	8	8	8	8	8
	TempMin	8	8	8	8	8
	Precip	8	8	8	8	8
	Water	8	8	8	8	8

Variables Entered/Removed[a]

Model	Variables Entered	Variables Removed	Method
1	Water, SoilDrain, Precip, TempMin[b]	.	Enter

a. Dependent Variable: AllCOProp

b. All requested variables entered.

Model Summary[b]

Model	R	R Square	Adjusted R Square	Std. Error of the Estimate
1	.852[a]	.727	.362	.0355716

a. Predictors: (Constant), Water, SoilDrain, Precip, TempMin

b. Dependent Variable: AllCOProp

ANOVA[a]

Model		Sum of Squares	df	Mean Square	F	Sig.
1	Regression	.010	4	.003	1.992	.299[b]
	Residual	.004	3	.001		
	Total	.014	7			

a. Dependent Variable: AllCOProp

b. Predictors: (Constant), Water, SoilDrain, Precip, TempMin

Coefficients[a]

Model		Unstandardized Coefficients		Standardized Coefficients	t	Sig.	95.0% Confidence Interval for B		Correlations			Collinearity Statistics	
		B	Std. Error	Beta			Lower Bound	Upper Bound	Zero-order	Partial	Part	Tolerance	VIF
1	(Constant)	-.216	.141		-1.530	.223	-.666	.234					
	SoilDrain	.018	.052	.148	.347	.751	-.147	.183	.491	.197	.105	.503	1.987
	TempMin	-.002	.004	-.263	-.545	.624	-.014	.010	.095	-.300	-.165	.393	2.545
	Precip	.007	.004	.851	2.017	.137	-.004	.019	.751	.759	.609	.512	1.954
	Water	.128	.154	.303	.832	.467	-.362	.618	.130	.433	.251	.685	1.460

a. Dependent Variable: AllCOProp

Coefficient Correlations[a]

Model			Water	SoilDrain	Precip	TempMin
1	Correlations	Water	1.000	-.428	.451	-.547
		SoilDrain	-.428	1.000	-.593	.677
		Precip	.451	-.593	1.000	-.666
		TempMin	-.547	.677	-.666	1.000
	Covariances	Water	.024	-.003	.000	.000
		SoilDrain	-.003	.003	.000	.000
		Precip	.000	.000	1.362E-5	-9.245E-6
		TempMin	.000	.000	-9.245E-6	1.416E-5

a. Dependent Variable: AllCOProp

Collinearity Diagnostics[a]

			Condition	Variance Proportions				
Model	Dimension	Eigenvalue	Index	(Constant)	SoilDrain	TempMin	Precip	Water
1	1	4.102	1.000	.00	.01	.00	.00	.01
	2	.712	2.400	.00	.00	.00	.00	.68
	3	.177	4.820	.00	.45	.00	.00	.00
	4	.006	26.745	.87	.00	.05	.34	.00
	5	.003	35.128	.13	.54	.95	.66	.31

a. Dependent Variable: AllCOProp

Residuals Statistics[a]

	Minimum	Maximum	Mean	Std. Deviation	N
Predicted Value	-.018881	.088155	.038800	.0379562	8
Std. Predicted Value	-1.520	1.300	.000	1.000	8
Standard Error of Predicted Value	.016	.036	.027	.007	8
Adjusted Predicted Value	-.060296	2.216835	.321053	.7690212	8
Residual	-.0313430	.0439452	.0000000	.0232871	8
Std. Residual	-.881	1.235	.000	.655	8
Stud. Residual	-1.309	1.697	-.198	1.077	8
Deleted Residual	-2.1682353	.0828935	-.2822528	.7649492	8
Stud. Deleted Residual	-1.631	6.895	.407	2.744	8

Mahal. Distance	.539	6.123	3.500	1.932	8
Cook's Distance	.000	742.869	93.309	262.463	8
Centered Leverage Value	.077	.875	.500	.276	8

a. Dependent Variable: AllCOProp

C7. Simple linear regression for adults and $\delta^{13}C$

Variables Entered/Removed[a]

Model	Variables Entered	Variables Removed	Method
1	CarbAvg[b]	.	Enter

a. Dependent Variable: SQRAdult

b. All requested variables entered.

Model Summary[b]

Model	R	R Square	Adjusted R Square	Std. Error of the Estimate
1	.011[a]	.000	-.125	.09496

a. Predictors: (Constant), CarbAvg

b. Dependent Variable: SQRAdult

ANOVA[a]

Model		Sum of Squares	df	Mean Square	F	Sig.
1	Regression	.000	1	.000	.001	.977[b]
	Residual	.072	8	.009		
	Total	.072	9			

a. Dependent Variable: SQRAdult

b. Predictors: (Constant), CarbAvg

Coefficients[a]

Model		Unstandardized Coefficients		Standardized Coefficients	t	Sig.
		B	Std. Error	Beta		
1	(Constant)	.305	.348		.878	.406
	CarbAvg	.001	.030	.011	.030	.977

a. Dependent Variable: SQRAdult

C8. Simple linear regression for children and $\delta^{13}C$

Variables Entered/Removed[a]

Model	Variables Entered	Variables Removed	Method
1	CarbAvg[b]	.	Enter

a. Dependent Variable: SQRChild

b. All requested variables entered.

Model Summary[b]

Model	R	R Square	Adjusted R Square	Std. Error of the Estimate
1	.726[a]	.528	.469	.14460

a. Predictors: (Constant), CarbAvg

b. Dependent Variable: SQRChild

ANOVA[a]

Model		Sum of Squares	df	Mean Square	F	Sig.
1	Regression	.187	1	.187	8.942	.017[b]
	Residual	.167	8	.021		
	Total	.354	9			

a. Dependent Variable: SQRChild

b. Predictors: (Constant), CarbAvg

Coefficients[a]

Model		Unstandardized Coefficients		Standardized Coefficients	t	Sig.
		B	Std. Error	Beta		
1	(Constant)	1.461	.361		4.048	.004
	CarbAvg	.096	.032	.726	2.990	.017

a. Dependent Variable: SQRChild

C9. Simple linear regression for adults and historic infection rates

Variables Entered/Removed[a]

Model	Variables Entered	Variables Removed	Method
1	historic[b]	.	Enter

a. Dependent Variable: SQRAdult

b. All requested variables entered.

Model Summary

Model	R	R Square	Adjusted R Square	Std. Error of the Estimate
1	.441[a]	.194	.079	.17984

a. Predictors: (Constant), historic

ANOVA[a]

Model		Sum of Squares	df	Mean Square	F	Sig.
1	Regression	.055	1	.055	1.690	.235[b]
	Residual	.226	7	.032		
	Total	.281	8			

a. Dependent Variable: SQRAdult

b. Predictors: (Constant), historic

Coefficients[a]

Model		Unstandardized Coefficients		Standardized Coefficients	t	Sig.
		B	Std. Error	Beta		
1	(Constant)	.131	.121		1.076	.317
	historic	.653	.502	.441	1.300	.235

a. Dependent Variable: SQRAdult

C10. Simple linear regression for children and historic infection rates

Variables Entered/Removed[a]

Model	Variables Entered	Variables Removed	Method
1	historic[b]	.	Enter

a. Dependent Variable: SQRChild

b. All requested variables entered.

Model Summary

Model	R	R Square	Adjusted R Square	Std. Error of the Estimate
1	.972[a]	.944	.926	.05665

a. Predictors: (Constant), historic

ANOVA[a]

Model		Sum of Squares	df	Mean Square	F	Sig.
1	Regression	.164	1	.164	50.998	.006[b]
	Residual	.010	3	.003		
	Total	.173	4			

a. Dependent Variable: SQRChild

b. Predictors: (Constant), historic

Coefficients[a]

Model		Unstandardized Coefficients		Standardized Coefficients	t	Sig.
		B	Std. Error	Beta		
1	(Constant)	-.037	.055		-.678	.546
	historic	1.790	.251	.972	7.141	.006

a. Dependent Variable: SQRChild

C11. Multiple linear regression, historic infection rates, four environmental variables

Descriptive Statistics

	Mean	Std. Deviation	N
historic	.162344	.1065606	9
SoilDrain	.7931522	.34295075	9
Elev	218.333	130.4329	9
Precip	49.556	6.4263	9
Water	.0408811	.03620578	9

Correlations

		historic	SoilDrain	Elev	Precip	Water
Pearson Correlation	historic	1.000	.400	.505	-.057	-.180
	SoilDrain	.400	1.000	.775	-.533	.388
	Elev	.505	.775	1.000	-.640	-.127
	Precip	-.057	-.533	-.640	1.000	.124
	Water	-.180	.388	-.127	.124	1.000
Sig. (1-tailed)	historic	.	.143	.083	.443	.322
	SoilDrain	.143	.	.007	.070	.151
	Elev	.083	.007	.	.032	.372
	Precip	.443	.070	.032	.	.376
	Water	.322	.151	.372	.376	.
N	historic	9	9	9	9	9
	SoilDrain	9	9	9	9	9
	Elev	9	9	9	9	9
	Precip	9	9	9	9	9
	Water	9	9	9	9	9

Variables Entered/Removed[a]

Model	Variables Entered	Variables Removed	Method
1	Water, Precip, Elev, SoilDrain[b]	.	Enter

a. Dependent Variable: historic

b. All requested variables entered.

Model Summary

Model	R	R Square	Adjusted R Square	Std. Error of the Estimate
1	.671[a]	.450	-.100	.1117662

a. Predictors: (Constant), Water, Precip, Elev, SoilDrain

ANOVA[a]

Model		Sum of Squares	df	Mean Square	F	Sig.
1	Regression	.041	4	.010	.818	.575[b]
	Residual	.050	4	.012		
	Total	.091	8			

a. Dependent Variable: historic

b. Predictors: (Constant), Water, Precip, Elev, SoilDrain

Coefficients[a]

Model		Unstandardized Coefficients		Standardized Coefficients			Correlations			Collinearity Statistics	
		B	Std. Error	Beta	t	Sig.	Zero-order	Partial	Part	Tolerance	VIF
1	(Constant)	-.418	.475		-.880	.429					
	SoilDrain	.188	.296	.606	.637	.559	.400	.303	.236	.152	6.582
	Elev	.000	.001	.313	.355	.741	.505	.175	.132	.176	5.670
	Precip	.009	.008	.521	1.058	.350	-.057	.468	.392	.567	1.762
	Water	-1.294	1.781	-.440	-.727	.508	-.180	-.341	-.269	.376	2.662

a. Dependent Variable: historic

Coefficient Correlations[a]

Model			Water	Precip	Elev	SoilDrain
1	Correlations	Water	1.000	-.182	.679	-.785
		Precip	-.182	1.000	.183	.189
		Elev	.679	.183	1.000	-.837
		SoilDrain	-.785	.189	-.837	1.000
	Covariances	Water	3.171	-.003	.001	-.413
		Precip	-.003	6.663E-5	1.080E-6	.000
		Elev	.001	1.080E-6	5.204E-7	.000
		SoilDrain	-.413	.000	.000	.087

a. Dependent Variable: historic

Collinearity Diagnostics[a]

			Condition	Variance Proportions				
Model	Dimension	Eigenvalue	Index	(Constant)	SoilDrain	Elev	Precip	Water
1	1	4.386	1.000	.00	.00	.00	.00	.01
	2	.391	3.349	.00	.00	.03	.00	.27
	3	.204	4.632	.01	.01	.04	.02	.07
	4	.015	17.260	.00	.92	.90	.00	.62
	5	.004	35.215	.99	.07	.02	.98	.04

a. Dependent Variable: historic

APPENDIX D

Spearman's correlation output from SPSS

D1. Spearman's correlation, adults and $\delta^{13}C$

Correlations

			AdLesProp	CarbAvg
Spearman's rho	AdLesProp	Correlation Coefficient	1.000	-.030
		Sig. (2-tailed)	.	.934
		N	20	10
	CarbAvg	Correlation Coefficient	-.030	1.000
		Sig. (2-tailed)	.934	.
		N	10	14

D2. Spearman's correlation, children and $\delta^{13}C$

Correlations

			SQRChild	historic
Spearman's rho	SQRChild	Correlation Coefficient	1.000	.829*
		Sig. (2-tailed)	.	.042
		N	17	6
	historic	Correlation Coefficient	.829*	1.000
		Sig. (2-tailed)	.042	.
		N	6	10

*. Correlation is significant at the 0.05 level (2-tailed).